MEASURING TEMPERATURES AND HEAT FLUX FROM DOLPHINS IN THE EASTERN TROPICAL PACIFIC: IS THERMAL STRESS ASSOCIATED WITH CHASE AND CAPTURE IN THE ETP-TUNA PURSE SEINE FISHERY?

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EXECUTIVE SUMMARY

Thermal stress can be associated with chase and capture in terrestrial mammals. Increased core temperatures in terrestrial mammals can cause maladaptive physiological changes and in extreme cases, even death. Hyperthermia may be most severe in pregnant mammals where blood flow to the uterus - required for adequate thermoregulation of the developing fetus - may be compromised. Decreased blood flow to, and concomitant increased temperatures at the fetus cause detrimental effects including low birth weight, retarded growth, skeletal and neurological developmental anomalies, and ultimately acute fetal stress and death. Thus, comparative studies demonstrate that severe hyperthermia can negatively impact both the health of the individual and its reproductive success.

Based on observations on terrestrial mammals, it is possible that dolphins undergoing a prolonged (20 minute- 2 hour), high-speed chase in the warm surface waters of the Easter Tropical Pacific (ETP) may be experiencing hyperthermic stress. This component of CHESS was designed to test the hypothesis that dolphins that are chased and captured in the ETP-tuna purse seine fishery suffer thermal stress. We used three complementary data sets to investigate the thermal biology of ETP dolphins: (1) deep core body temperatures, (2) infrared thermographic images, and (3) thermal data logger records.

The results of this study suggest the following interpretation of the thermal response of dolphins to chase and capture. A dolphin's thermal response begins during the helicopter phase of the chase. Chased dolphins appear to respond by increasing the rate of heat dissipation to the environment. Prolonged chase times (75 minutes or longer) result in measurably higher skin surface temperatures of dolphins. These elevated surface temperatures suggest relatively high rates of heat delivery to the skin's surface *via* blood flow. Dolphins that have experienced chases less than 75 minutes in length do not appear to maintain elevated levels of heat flux and high skin surface temperatures once the chase ends and the animals are within the net corral. This result may be, in part, because the animals in this study did not appear to experience any impediment to free-swimming, and, thus, to convective heat dissipation, within the net corral. Extended chase durations and capture times within the net were not statistically associated with elevated deep body temperatures in this study. One of the 48 individuals, though, displayed an unusually high body temperature of 37.9°C. Because there was no evidence from blood diagnostic tests to confirm illness, physiological or exertional stress is the more likely explanation of this individual's elevated body temperature.

INTRODUCTION

Thermal stress can be associated with chase and capture in terrestrial mammals (e.g. Antognini *et al.* 1996, Beringer *et al.* 1996). Increased core temperatures in terrestrial mammals can cause maladaptive physiological changes and in extreme cases, even death (e.g. Taylor *et al.* 1972). Hyperthermia may be most severe in pregnant mammals where blood flow to the uterus - required for adequate thermoregulation of the developing fetus - may be compromised by shunting arterial blood flow to either (1) exercising locomotory muscles and/or (2) skin to dump excess heat at the body surface (reviewed in Bell and Hales 1986). Decreased blood flow to, and concomitant increased

temperatures at the fetus cause detrimental effects including low birth weight, retarded growth, skeletal and neurological developmental anomalies, and ultimately acute fetal stress and death (reviewed in Rommel *et al.* 1993).

Based on observations on terrestrial mammals, it is possible that dolphins undergoing a prolonged (20 minute- 2 hour), high-speed chase in the warm surface waters of the ETP may be experiencing hyperthermic stress. Like other mammals, dolphins can regulate their body temperature by controlling the flow of blood to the body surface (e.g. Scholander and Schevill 1955). The dolphin's uninsulated dorsal fin and flukes function as "thermal windows" across which excess body heat is dumped to the environment (reviewed in Pabst *et al.* 1999). Blood routed through veins on the surfaces of these extremities is cooled by heat transfer to the surrounding water and, when returned to the body core, can be used to achieve whole body cooling (Scholander and Schevill 1955). We have discovered that most of the blood routed through these superficial veins, though, is returned to the deep body *via* a venous plexus that is, in turn, juxtaposed to the arterial supply to the reproductive tissues (Rommel *et al.* 1992). Thus, dolphins possess a reproductive countercurrent heat exchanger (CCHE) that is in a position to cool the uterus and, hence, the developing fetus in females (Rommel *et al.* 1993, Pabst *et al.* 1998, Rommel *et al.* 1998) and the cryptic testes in males (Rommel *et al.* 1992).

Deep body temperatures of male bottlenose dolphins (taken with a custom-built colonic probe housing a linear array of thermocouples) demonstrate that temperatures in the region of the colon flanked by the CCHE are generally cooler than temperatures measured in front of or behind it (Rommel *et al.* 1994). Manipulation of the surfaces of the dorsal fin and flukes causes increases in body temperature at the region of the CCHE. Controlled and moderate surface swimming causes decreases in deep body temperatures in the region of the CCHE, but increases in body temperatures at other positions along the colon (Pabst *et al.* 1995). We hypothesize that these temperature changes are caused by changes in the temperature and/or the flow rate of blood returning from the dorsal fin and flukes (Pabst *et al.* 1995). Thus, the vascular design of the dorsal fin and flukes contributes to thermoregulation of both the whole body and the reproductive systems.

Although the CCHE in bottlenose dolphins has an increased ability to regulate the temperature of the testes during controlled and moderate exercise, we do not understand the effects of prolonged chase and subsequent capture on any aspect of thermoregulation in dolphins. In terrestrial mammals, high temperatures at the testis can block sperm production and maturation, and elevated temperatures at the uterus can cause fetal distress.

Thus, comparative studies demonstrate that severe hyperthermia can negatively impact both the health of the individual and its reproductive success. Our component of CHESS was designed to test the hypothesis that dolphins that are chased and captured in the ETP-tuna purse seine fishery suffer thermal stress. We used three complementary data sets to investigate the thermal biology of ETP dolphins: (1) deep core body temperatures, (2) infrared thermographic images, and (3) thermal data logger records. We briefly discuss below the hypotheses tested with each data set.

Deep Core Body Temperatures

Dolphins targeted by the tuna fishery can undergo prolonged chases in the warm surface waters of the ETP, and, thus, may experience elevated temperatures. Chased dolphins are then captured and temporarily held within the net corral. It is possible that captured dolphins may also experience increased body temperatures, if the capture process decreases their ability to swim freely and, thus, to dissipate excess body heat *via* convection.

If chase and capture cause increases in deep body temperatures, we hypothesized the following.

(a) Dolphins that had been chased for longer periods of time would have higher deep body temperatures than those that had experienced shorter chase times.

(b) Dolphins that had been held in the net corral for longer periods of time would have higher deep body temperatures than those that had experienced shorter capture times.

Infrared Thermography

Infrared thermal images permitted us to measure surface body temperatures of dolphins within the net corral. Because dolphins use their dorsal fins as thermoregulatory surfaces (Scholander and Schevill 1955, Hampton *et al.* 1971, McGinnis *et al.*1972, Hampton and Whittow1976, and Noren *et al.*1999, Williams *et al.* 1999, Pabst *et al.* 2001), we hypothesized that increases in deep body temperatures during chase should be reflected by increased surface temperatures at the dorsal fin. Infrared data collected before the CHESS cruise suggested that the surface temperature of the fin increased with exercise, as dolphins shunted warm blood to the fin's surface to dissipate excess body heat (Pabst *et al.* 2001). In dolphins that were held stationary after exercise, fin temperature also increased as time after exercise increased, presumably because the animal was not able to use convection to dissipate excess body heat. Thus, we hypothesized the following.

(a) Dolphins that had been chased for longer periods of time would have higher surface temperatures than those that had experienced shorter chase durations.

(b) Dolphins that had been encircled within the net corral for longer periods of time would have higher surface temperatures than those that had been encircled for shorter periods, if that encirclement constrained their ability to swim freely.

Thermal Data Logger Deployments

Thermal data loggers permitted us to measure heat flux and temperatures from the dorsal fins of free-swimming animals. Heat flux is the amount of heat energy (Watts) per unit area (m^2) that moves across a thermodynamic barrier, in this case heat from the blood moving across the surface of the dorsal fin. The thermal data logger offered us the opportunity to measure changes in the thermal interactions between a dolphin and its environment during chase and capture. Because dolphins use their dorsal fins as thermoregulatory surfaces we hypothesized that increases in deep body temperatures during chase should be reflected by increased heat flux values (and temperature) at the

dorsal fin. Heat flux and skin surface temperatures collected before the CHESS cruise suggested that at the fin, both values increased with exercise, as dolphins shunted warm blood to the fin's surface to dissipate excess body heat (Pabst *et al.* 2001). The relationship between body temperature and heat flux is, though, complex and our recent captive animal work suggests that there is a latency period between the finish of exercise and the highest observed heat flux rates. Therefore, the most extreme thermal effects (*i.e.* highest heat flux values) may occur after the chase is over. We hypothesized that if chase and capture cause increases in deep body temperatures, we should observe the following.

(1) Dorsal fin heat flux values and skin surface temperatures would be higher in a dolphin during and after a chase than those recorded before the chase began.

(2) A dolphin that had undergone chase and capture would have higher heat flux and skin surface temperatures directly after initial capture than those measured at later times.

(3) A dolphin that experiences a prolonged chase would maintain elevated heat flux and skin surfaces temperatures for a prolonged period while within the net corral.

An independent scientific peer review of this work was administered by the Center for Independent Experts located at the University of Miami. Responses to reviewer's comments can be found in the Appendix.

METHODS

Deep Core Body Temperatures
Our goal was to determine whether there was an effect of chase duration and/or length of capture time in the tuna net on deep body temperatures of dolphins. Deep body temperatures were collected from spotted dolphins (*Stenella attenuata*) while they were being processed in the rafts. Temperatures were recorded using an Omega veterinary colonic probe (copper constantan thermocouple sensor), downloaded and stored every 30 seconds on a FLUKE 54 II digital thermometer. Deep body temperatures were measured at an insertion depth of approximately 20 cm. Only those dolphins that had core temperatures collected for at least 2 minute were included in the analyses.
For analyses, deep body temperatures from all sets were pooled and the time of temperature measurement, relative to the chase or capture event, was calculated. Chase duration, defined here as the amount of time between "helicopter over the dolphins" and "net let go", was determined for each set. Standard regression analysis was performed on the pooled data to examine the relationship between deep body temperatures and chase duration. We also divided the helicopter chase durations into 15-minute segments (see Table 1), and compared mean deep body temperatures across time blocks. Binning the data allowed increased samples sizes within each discrete time block. One-way ANOVAs were used to determine whether there were significant differences in mean temperature differentials across the different chase durations. The Tukey-Kramer Honestly Significant Difference test was subsequently performed to determine which

mean temperature differentials were significantly different from each other (JMPIN 4.03, SAS Institute, Inc.).

Table 1. Chase durations (in minutes) from helicopter over dolphins to let go and from first speedboat in water to let go (see also thermal imaging section).

		Helicopter	Speedboat
1		0-15	0-5
2		16-30	6-10
3		31-45	11-15
4		46-60	16-20
5		61-75	21-25
6		76-90	
7		91-105	
8		106-120	

Regression analysis was performed to examine the effects of capture duration (*i.e.* the amount of time elapsed since net let go) on deep body temperatures (Microsoft Excel). An additional regression analysis was performed to investigate the relationship between deep body temperature and total body length. Deep body temperatures of males *vs*. females were also compared. For recaptured individuals, serial measurements of deep body temperatures were graphed to investigate whether there was a cumulative effect of chase and capture on measured body temperatures. To determine whether there was an effect of amount of time the dolphin spent in the raft on deep body temperatures, mean deep body temperatures for females and for males were plotted against time in raft.

Infrared Thermography

Thermal images were collected using a FLIR systems AGEMA 570 infrared camera with an attached 12° FOV lens. The lens was covered with a single layer of clear Saran Wrap to protect it from salt spray. Images were stored on a SanDisk flash memory card and downloaded after each set to a laptop computer for analysis.

For each set, thermal images of dolphins were collected as soon as possible after the net was released and encirclement begun. Most images taken were of spotted dolphins, although spinner dolphins (*Stenella longirostris orientalis*) were also imaged in sets on mixed-species schools. Images were then collected throughout the set and at backdown. To obtain images as soon as possible after encirclement, the camera and operator were launched in the first RHIB, which would enter the area inside the net once the net was closed. The RHIB circled the area inside the net allowing the camera operator to collect as many thermal images of captured dolphins as possible until just prior to tiedown. At that point in time, the RHIB left the area inside the net and relocated to an advantageous position on the outside of the net to continue taking thermal images.

During backdown, the RHIB stationed itself to the side of the channel and thermal images were taken as animals exited the net. Infrared images were taken opportunistically, and the individual identities of imaged dolphins were not known. Thus, during any given set, multiple images of an individual dolphin could have been collected.

Thermal images were analyzed using FLIR systems ThermaCAM Researcher 2001 software. The time of each thermal image was recorded by the FLIR camera and retrieved during image analysis. The thermal camera's time was synchronized to GPS time, the standard used for all CHESS project data. Surface temperatures for dolphin dorsal fins and bodies and for the water's surface were measured in each high quality image (*i.e.* animals in focus). Skin surface temperatures of the dorsal fin were measured inside an oval shaped area at the center of the fin (Figure 1), while body temperatures were measured inside a rectangular area on the maximum exposed surface of the body (see Figure 2). Inside each area, a maximum, minimum and mean temperature was calculated. The integrated mean temperature for each area was used as the surface temperature in all analyses.
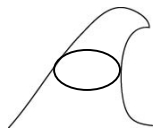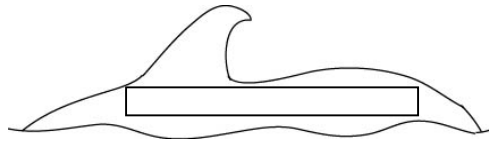
Figure 1   **Figure 2**

Water surface temperatures were determined inside three circular shaped areas on each image. These circular shaped areas were placed on each image to encompass the maximum visible area of water surface. Because the distance from the photographer to the dolphin changed from image to image, the total water surface area sampled also varied across images within a set. This effect, though, was similar across all sets. The mean water temperature inside each area was determined and the overall mean of the three circles was calculated and used as the water temperature in all analyses for that image.

The infrared camera measured only the temperature of the very surface of the water. Interestingly, these temperatures were often a few degrees warmer than more standard surface water temperatures, which were measured at a few centimeters depth. To ensure that surface mixing during higher sea states was not affecting temperatures recorded by the infrared camera, mean water surface temperature was also compared across Beaufort sea states.

Because Saran Wrap was placed over the lens in all images collected during sets, a calibration equation was needed to correct for any effects of Saran Wrap on measured surface temperatures. The range of temperatures (dorsal fin, body or water) collected from the thermal images in the ETP was 28.5-36.1^{o}C. Thus, objects in this temperature range were imaged in the lab at UNCW both with and without the lens covered in Saran Wrap. A total of 29 measurements were taken from these objects. Linear regression analysis was performed to establish the relationship between measured temperatures with and without Saran Wrap (see Figure 3). The equation for this linear fit was:

without Saran Wrap temp = -13.13789 + 1.4969785 (with Saran Wrap temp)

This equation was used to correct for the effects of Saran Wrap in all temperatures from images collected during tuna set operations.
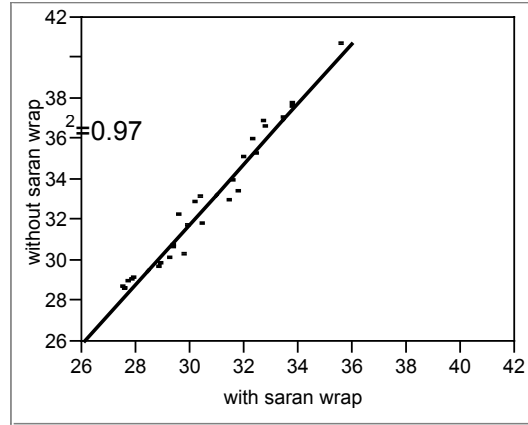

Figure 3

In initial analyses, average dorsal fin and body surface temperatures were graphed against the corresponding water surface temperatures. Regression lines were fit to these graphs to examine any effect of water surface temperature on skin surface temperature (JMPIN 4.03, SAS Institute, Inc.). For all remaining analyses, images from all sets were pooled and the time of each image, relative to the chase or capture event, was calculated. Thus, we used a similar approach with skin surface temperatures as we did with deep body temperatures.

Our goal was to test whether there was an effect of chase and/or capture duration in the tuna net on the surface temperatures of dolphin dorsal fins and bodies. To examine the relationship between chase duration and skin surface temperatures, the amount of time between (1) helicopter over the dolphins and net let go (total chase duration) and (2) first speedboat in the water and net let go were determined for each set. The total chase durations were divided into 15-minute segments, while speedboat chase durations were divided into 5-minute segments (see Table 1). The dorsal fin and body temperature differentials (T_{skin} - T_{water}) were compared across time blocks. Binning the data allowed increased samples sizes within each discrete time block. One-way ANOVAs were used to determine whether there were significant differences in mean temperature differentials across the different chase durations. The Tukey-Kramer Honestly Significant Difference test (JMPIN) was subsequently performed to determine which mean temperature differentials were significantly different from each other (see Table 3 for sample sizes). Standard regression analyses (JMPIN) were also performed on the pooled data to examine the relationship between skin surface temperatures and total chase duration and the amount time spent within the net corral.

We also compared dorsal fin and body temperatures of dolphins at the extremes of the capture process. All animals imaged within 30 minutes of let go (n=10 images for dorsal fin temperature, n=7 images for body temperature) were pooled and compared, using one-way ANOVAs (JMPIN), to the animals imaged within 20 minutes of

backdown complete (n=76 images for dorsal fin temperature, n=46 images for body temperature).

Additional analyses were performed comparing the relationship between skin surface temperature measured while an animal was on a raft with its skin more exposed to air (dorsal fin, n=10 images; body, n=6 images) and skin surface temperature measured while an animal was in the water, and its skin exposed to air only during surfacing events (dorsal fin, n=422 images; body, n=202 images). While in the raft, dolphins were partially submerged in about 10-15cm of water. Their dorsal fins were exposed to air although they were occasionally splashed with water during the handling process. Because water surface temperature could not be obtained from images of animals in a raft, average skin surface temperature was used for these comparisons rather than the temperature differential. For the dorsal fin surface temperatures, animals without tags (n=2 images) were compared to those with tags (either carrying a tag or with a tag recently removed; n=8 images) in air and in water. As there were no significant differences between tagged and untagged animals (Tukey-Kramer HSD test, JMPIN) all images of dorsal fins in air were pooled and compared to images of fins in water. (Note: The small sample sizes suggest that our ability to detect differences between tagged and un-tagged animals is very low.) A one-way ANOVA and Tukey-Kramer HSD test were used to test for significant differences between the mean average skin surface temperatures of dolphins in air and dolphins in water (JMPIN).

Table 3. Sample sizes used for the analyses of the effect of chase duration (helicopter over dolphins to let go) on skin surface temperature.

	Dorsal Fin Surface Temperature		**Body Surface Temperature**	
Chase Duration (helicopter)	# of sets containing images used	# of images used	# of sets containing images used	# of images used
0-15	2	14	1	2
16-30	3	59	3	29
31-45	5	82	3	58
46-60	2	36	2	11
61-75	1	19	1	2
76-90	1	128	1	45
91-105	0	0	0	0
106-120	1	84	1	55

Thermal Data Logger Deployments

The heat flux transducer (Vatell episensor B02, Vatell Corp. Christensburg VA) produces a weak signal (0-0.1 mV) at frequencies from dc out to about 1000 Hz. Therefore we designed a low noise differential dc amplifier with a stable zero and flat response curve that would amplify this signal to 0-2.5 volts. Each amplifier had a gain of approximately 2500x. These voltages were then recorded using a modified HOBO 4-channel data logger (Onset Computer Corp. Pocasset, MA). The package also contained a HOBO type T thermocouple data logger (Onset Computer Corp, Pocasset, MA). The probe from this device was attached to the face of the heat flux disk, which allowed heat flux and temperatures to be collected from the same site on the dorsal fin.

The thermal logger was attached to the dolphin's dorsal fin using a thermo-plastic saddle that had been vacuum molded to fit the contours of the "average" *Stenella attenuata* dorsal fin. The saddle was lined with open cell foam (1/8"). Two 7.0mm delrin pins were placed through the dorsal fin after holes had been cut using a sterilized cork-boring tool. These pins were secured using Mg nuts and steel washers. In the event that the animal was not recaptured this galvanic linkage would corrode and break allowing the package to release from the dolphin. We inserted foam washers between the saddle and the dorsal fin at each pin site. These washers served to raise the saddle off of the dorsal fin and allow water flow, which was done in order to minimize any insulative effects caused by the saddle. The saddle plastic was constructed from 1/8" modified polyethylene. The plastic had been molded around two syntactic foam (Emerson and Cuming, Canton MA) blocks (approx. 10.5 x 7.0 x 2.5 cm) that had been previously formed into a hydrodynamic shape. These blocks formed two pockets on each side of the saddle that were partially hollowed out to accommodate the electronic packages. On the right side of the saddle we attached a VHF radio (Holohil, Carp, ON) and Mk-8 time-depth recorder (TDR) (Wildlife Computers, Woodenville WA). The VHF transmitters allowed us to radio-track the animals throughout each deployment and recover the packages once they had come off. The TDR allowed us to record the diving and velocity behavior, and water temperature. On the left side we attached the logger electronic package. The heat flux disk (2.5 x 2.5 cm) was attached to a short spring and pushed up gently against the fin when the saddle was in place. The relative position of the HF disk on each dorsal fin was different because of individual variation in dorsal fin size and shape. A short wire (5 cm) connected the HF disk to the electronic package. Skin temperatures were recorded using a thermocouple as described above. Water temperature data were collected using a thermistor on the TDR.

Because the attachment technique we used was invasive, the tagging process likely thermally affected the fin. Thus, thermal data collected during these deployments might be biased due to inflammatory processes, as well as other tag effects such as enhanced fin insulation, increased drag, etc. To determine the potential magnitude of tag-attachment effect, we compared the heat flux values from this study to those collected from Sarasota bottlenose dolphins wearing similar thermal loggers attached *via* less invasive suction cups.

RESULTS

Deep Core Body Temperatures

We collected 55 deep body temperature records from 48 individuals (25 females and 23 males) (see Table 2). One individual (D29) was sampled three times during three separate captures, and four individuals (D34, D227, D42, D63) were sampled during two capture events. All other individuals were sampled once. Two individuals, D17 and D47, had deep body temperatures that were considerably lower than all other dolphins. An outlier analysis demonstrated that their temperatures were outside the 99% confidence limit for the data set. Thus, these individuals were excluded from group analyses. D67 was also excluded because his deep core temperature was recorded 8 minutes, rather than 2 minutes, after probe insertion. D42 was excluded from those analyses that required data on chase duration, as this information was not recorded for this individual. D42 was,

though, included in analyses that investigated the relationships between deep body temperatures and (1) the amount of time spent within the net corral, (2) sex and (3) total body length.

Regression analysis demonstrated a weak (adjusted r^2=0.0976), but significant (p=0.0146), negative relationship between deep body temperatures and total chase duration (Figure 4). Interestingly, this relationship appeared to be due to females. When each sex was analyzed separately, there was a significant relationship between total chase duration and deep body temperatures for females (adjusted r^2= 0.1758; p=0.0169), but not for males (adjusted r^2= -0.0287; p= 0.5554) (Figure 5).

ANOVAs of deep body temperatures relative to binned time blocks of chase duration (see Table 1) revealed no detectable relationship for both sexes combined (n=51; F=1.88, df=50; p=0.1061), for males (n=24; F=1.20; df=23; p=0.3512), or for females (n=27; F=1.55; df=26; p=0.2166) (Figures 6 and 7). Because this analysis required distributing individuals into "bins" of time after the chase event, the small sample size of individuals within each bin, especially for the longer chase durations, suggests that caution is warranted when interpreting these results. The length of time in the net corral was also not related to deep body temperatures in either sex (adjusted r^2= -0.0115; p=0.5307) (Figure 8).

There were no differences between mean deep body temperatures for females and males (n=52; F=0.69; df=51; p=0.4118) (Figure 9). Additionally, deep body temperatures of all dolphins were not correlated to total body length (adjusted r^2= -0.0105; p=0.4955) (Figure 10). Separate analyses for each sex also demonstrated no significant relationship between deep body temperature and total body length.

Serially taken deep body temperatures of recaptured dolphins were generally similar across sample dates (Figure 11). Four out of six animals displayed minor decreases. D29 displayed a slight increase in deep body temperature across successive capture events. Interestingly, D29's second capture was preceded by one unsuccessful capture attempt, and her third capture attempt was preceded by two unsuccessful attempts. One of the recaptured dolphins, D67, (not shown in Figure 11) appeared to have a deep body temperature that was elevated by a degree, but the second temperature was recorded at 8, rather than 2, minutes after probe insertion.

Both male and female dolphins generally displayed continuously increasing deep body temperatures the longer they were held in the processing raft. Of the 23 deep body temperatures measured in females, 16 (70%) increased, five decreased and two remained unchanged. Figure 12 illustrates examples of individual trends in females. Figure 13 demonstrates that although the deep body temperatures of most females increased over time, there was no obvious trend in mean body temperatures as time in raft increased. This result is likely due to the large range of individual variation in deep body temperatures. Of the 23 deep body temperatures measured in males, 17 (74%) increased, three decreased, and three remained unchanged. Males generally showed similar patterns to those observed in females (Figures 14 and 15).

Infrared Thermography

We collected 623 thermal images during the CHESS cruise. Of these, 343 were of sufficient quality to be analyzed. Each image could contain multiple dorsal fins and/or bodies, which increased the total "n" for many analyses. Thermal images were only

collected in Beaufort sea states 2 and 3. Although the highest sea surface temperatures were measured in Beaufort sea state 2, there were no significant differences between water surface temperatures measured across sea states. There was, though, a strong linear relationship between water temperatures and skin surface temperatures (dorsal fin: adjusted r^2=0.9218; p<0.001; body: adjusted r^2=0.9249; p<0.001) (Figure 16). Recall that the temperatures measured by the infrared camera were only those of the very surface of the water, and that these temperatures were often a few degrees warmer than more standard measures of surface water temperatures.

The one-way ANOVAs suggested that there was a significant effect of the total preceding chase time on the temperature differential measured between skin and water (dorsal fin: n= 422; F=20.93; df=421; p<0.0001; body: n=202; F=16.53;df=201; p<0.0001). Animals that had been chased for over 75 minutes had higher dorsal fin temperature differentials than those chased for less time, except for those that had been chased for 30-45 minutes (Figure 17a). Interestingly, animals that had been chased for 30-45 minutes had higher dorsal fin temperature differentials than those that had been chased for 45-75 minutes. A similar pattern was seen in body temperature differentials (Figure 17b). Animals that had been chased for over 75 minutes had higher body temperature differentials than those chased for 0-60 minutes. We had a very small sample size (n=2) for animals chased for 60-75 minutes, and there were no significant differences between this chase duration and any others. Because the samples sizes for these analyses were highly variable (see Table 3), and the data may have included multiple infrared images from the same individual, caution should be exercised when interpreting significance levels. Regression analyses, though, also demonstrated weak but significant, positive relationships between skin surface temperatures and total chase duration for both the dorsal fin (adjusted r^2=0.0633; p<0.001) and the body (adjusted r^2=0.1800; p<0.001) (Figure 18).

The one-way ANOVAs also suggested that there was a significant effect of the preceding speedboat chase time on the temperature differential measured between skin and water (dorsal fin: n= 422; F=18.99; df=421; p<0.0001; body: n=202; F=13.89; df=201; p<0.0001). The relationship between skin surface temperatures and speedboat chase times, though, were different than those seen with total chase times. Animals that had been chased by speedboats for 0-5 minutes and for 10-15 minutes had higher dorsal fin temperature differentials than those chased for 5-10 minutes and those chased for 20-25 minutes (Figure 19a). A similar pattern was seen in body temperature differentials (Figure 19b). In this case, animals that had been chased by speedboats for 0-5 minutes and for 10-15 minutes had higher body temperature differentials than those chased for 5-10 minutes. We had a very small sample size (n=2) for animals chased for 20-25 minutes, and there were no significant differences between this chase duration and any others. Regression analysis demonstrated no relationship between speedboat chase durations and the temperature differential measured between skin and water (dorsal fin: adjusted r^2= -0.0022; p=0.8008; body: adjusted r^2=0.0004; p=0.2967) (Figure 20).

There appeared to be no relationship between surface temperature differentials and length of time spent within the net corral. Neither the dorsal fin temperature differential (adjusted r^2= 0.0020; p=0.1769) (Figure 21) nor the body temperature differential (adjusted r^2= -0.0020; p=0.3969) (Figure 22) varied with time in the net. Temperature differentials were also examined at the extreme ends of the capture process.

Thermal images of dorsal fins (n= 86; F=0.46; df=85; p=0.4973) (Figure 23a) and bodies (n=53; F=0.0001; df=52; p=0.9917) (Figure 23b) taken within 30 minutes of net out showed temperature differentials similar to those of animals imaged within 20 minutes of the backdown procedure being completed.

Dorsal fin surface temperatures of dolphins temporarily restrained in the processing rafts were significantly higher than those of animals swimming in the net compass, whether the animals had been tagged or not (Figure 24). Dorsal fin surface temperatures of tagged and untagged animals did not differ from each other when measured in air and in water (Figure 24a). Thus, in both air and water all tagged and untagged fins were pooled for further analyses. The mean fin temperature measured in air was significantly higher (by 3.4^{o}C) than those measured in water (n=432; F=72.92; df= 431; p<0.001) (Figure 24b). Similarly, the mean body temperatures measured in air was significantly higher (by 3.4^{o}C) than those measured in water (n=208; F=37.25; df=207; p<0.001) (Figure 25).

Thermal Data Logger Deployments

During CHESS the thermal logger was deployed on two occasions. The first deployment (D63) occurred on September 19 and was on a 190 cm male spotted dolphin. This deployment was 23 h 20m in duration. During deployment, the TDR dislodged from the saddle and was lost. Therefore, only heat flux and temperature were recorded. The second deployment (D227) occurred on September 22 and was on a 204 cm male spotted dolphin. This deployment was 76 hours in duration. From this dolphin we collected complete TDR and partial heat flux records. About 8 hours into the deployment there was a malfunction with the heat flux disk, which caused it to stop collecting data. It returned to normal operation during the final 6 hours of the deployment. The memory of the logger was limited to 72 hours of data collection therefore the tag turned off at 12 noon, about 4 hours before the animal was recaptured, during its third recapture attempt. Despite the malfunction we were able to collect a high quality data record immediately following release as well as during an initial, unsuccessful recapture attempt of D227 that occurred on September 25 between 07:40 and 10:20. Although this record is not the final capture event for D227, it represents a complete thermal record of the unsuccessful capture attempt, from time preceding the initiation of the helicopter chase, to virtual completion of the set. D227 was enclosed in the net compass during this event, but subsequently escaped.

The entire heat flux and temperature records from both deployments are shown in Figures 26-27. Figure 28 illustrates the edited record obtained from D227 after removing the time blocks during which the pack was inoperative. Details of the beginning and end of each heat flux/temperature record are shown in Figures 29-32. Generally, heat flux values were lowest immediately after the dolphins were released from the processing raft back into the net. These values increased markedly after backdown and escape (Figures 29 and 31).

During subsequent chase and capture, heat flux values increased quickly during the helicopter chase but sharply decreased as soon as the chase ended and the animals were inside the net corral (Figures 30 and 32). Skin temperatures decreased during and after capture in D63 but did not change in D227 (Figures 30 and 32). Examination of the

velocity record from the initial chase and capture attempt that was made on D227 revealed that there was no direct relationship between velocity and heat flux (Fig. 33).

We tested whether there were differences in the heat flux and skin temperature values before, during and after chase using one-way analysis of variance (SPSS 10.0 2000). We selected a five-minute interval at the following marks: immediately prior to helicopter take-off, immediately following the first sighting of animals within the net corral, and immediately after the net had been closed. These intervals are labeled pre, during and post in Figures 30 and 32. Post-hoc comparisons were made using Tamhane's T2 test since variances were not equal. Significant differences ($p<0.001$) in mean heat flux values were found between all time periods tested except pre- and during chase in D63. Notice that for both dolphins, heat flux values were elevated during the helicopter chase periods, but had decreased during the speedboat chase periods. On both deployments, heat flux values after the chase were significantly lower than those recorded before the chase began. Significant decreases were also documented in skin temperature in D63 during this time period, but not in D227.

DISCUSSION

We used three complementary data sets to investigate the thermal responses of dolphins to chase and capture in the ETP-tuna purse seine fishery: (1) deep body temperatures of captured dolphins, (2) dorsal fin and body surface temperatures of dolphins swimming in the net corral, and (3) dorsal fin heat flux values and skin surface temperatures of free-swimming dolphins. Each approach permitted us to collect relevant thermal data, but each had its limitations. Deep body temperatures offered us the most direct measure of the thermal status of individuals, but required handling the dolphins in the raft. Thus, the experimental procedure itself may have biased the results. This approach also sampled a relatively small number of individuals (n=48). Infrared thermal images permitted measurements of surface temperatures of hundreds of swimming corralled dolphins, but these images could not be correlated to any individual. Thus, multiple images of a single individual may exist within each set's samples, complicating statistical analyses of this otherwise robust data set. Thermal data loggers permitted us to measure the thermal responses of free-swimming individuals during chase and capture, but only two individuals were sampled. The combined results of these approaches, though, offer us insights into the thermal biology of chased and captured ETP dolphins.

Deep Core Body Temperatures
We hypothesized that if chase and capture posed a thermal threat to ETP dolphins, animals that had been chased and/or held within the net corral for longer periods of time would have higher body temperatures than those that had experienced shorter chase and/or capture times. Regression analyses suggested that there was no detectable relationship between chase duration and deep body temperatures for males and a weakly negative relationship for females (Figure 4 and 5). That is, the longer the chase, the lower the body temperatures measured in females.

If the deep body temperatures measured in ETP dolphins were being influenced by the CCHE, and if the CCHE functioned in chased spotted dolphins as it does in moderately exercising bottlenose dolphins (Pabst *et al.* 1995), then we might expect to

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see decreased deep body temperatures after exercise. We do not, though, know the position of our colonic probe, relative to the position of the CCHE, for any ETP dolphin due to a lack of morphological data. If the CCHE were influencing deep body temperatures, our results might suggest enhanced CCHE function in females, relative to males. Alternatively, the CCHE could be differentially affecting temperatures measured in females if there was a systematic difference in colonic probe placement, relative to the CCHE, between the sexes. If this were the case, we would expect to observe sex-based differences in total body length, as the probe was uniformly inserted to a depth of 20 cm in all dolphins. There was, though, no difference in mean body length of females *vs*. males (n=51, F=0.4076; df=50, p=0.5261), and there was also no relationship between total body length and deep body temperatures across both sexes (Figure 10). Thus, the difference in the relationship between female and male deep body temperatures and chase duration is not clear.

An alternative statistical approach, that of using ANOVAs of binned time blocks, demonstrated no detectable effect of chase duration on deep body temperatures measured for the individuals in this study (Figures 6 and 7). The small sample size of individuals within each time bin, especially for the longer chase durations, though, limits the power of this data set to detect differences. Furthermore, regression analysis demonstrated no relationship between deep body temperatures and the time spent within the net corral. Thus, in general, there does not appear to be a positive relationship between deep body temperatures of individuals sampled in this study and either the length of the chase they experienced, or the amount of time they spent within the net corral.

Three individuals displayed temperatures that were noticeably different from those of most dolphins. D17, a male, and D47, a female, displayed temperatures that were dramatically lower than other dolphins (outside the 99% confidence limit). These low temperature measurements may simply be artifactual – they may have occurred if the colonic probe was not inserted to the full 20 cm depth in these individuals. Although we cannot entirely rule out this possibility, we have no evidence from the continuous temperature records of these dolphins that this was the case. Our previous observations in wild bottlenose dolphins suggest that some pregnant females have considerably lower temperatures than those of other individuals (Pabst *et al*., unpublished data). D47 had the third highest progesterone level measured during this study (St. Aubin, 2002), which although not conclusive, may suggest that this individual was in an early stage of pregnancy. Of the two females with the highest progesterone levels (St. Aubin, 2002), D12 had a relatively low temperature (36.4°C) and D14 had a relatively high temperature (37.4°C). Thus, there is no clear relationship between deep body temperature and reproductive status in females sampled in this study.

It is interesting to note that D17, a male with a very low body temperature, also had the highest white blood cell count of any dolphin on its first capture (St. Aubin, 2002). His extremely high eosinophil and lymphocyte counts suggest a diagnosis of concurrent heavy parasitism and non-specific antigenic stimulation (St. Aubin, 2002). We do not know if there is a causal relationship between these two extreme measures in D17.

D61, a female, displayed an unusually high body temperature of 37.9°C, suggesting that this individual was potentially ill or physiologically or pathologically stressed. Blood diagnostic tests did not reveal any specific abnormalities to confirm

illness (St. Aubin, 2002), leaving physiological or exertional stress as the more likely explanation of this individual's elevated body temperature. Whether this exertional stress was due to the chase and capture by the tuna vessel, or by extended pursuit times within the net by the research swimmers (Scott, M.D., pers. comm.) is not known.

The required handling of the animals may have biased the temperature data gathered in this study. We can, though, compare the range of deep body temperatures measured from ETP dolphins with those of wild bottlenose dolphins (*Tursiops truncatus*) from Sarasota, FL (Pabst *et al.* 2001). The water temperatures for both environments are similar, ranging from 27.8-31.9 $^{\circ}$C in Sarasota and from 27.6- 29.8°C in the ETP (as measured from the *McArthur* during sets). Deep body temperatures of bottlenose dolphins in Sarasota, FL, taken during the summers of 2000 and 2001, ranged from 35.8°C to 37.7 $^{\circ}$C. Of 37 wild bottlenose dolphins investigated, 15 (40%) had temperatures that were higher than 37°C and 22 (60%) had temperatures lower than 37°C. In ETP dolphins, 27 (50%) had temperatures equal to or greater than 37°C, and 27 (50%) had temperatures less than 37°C. Thus, deep body temperatures are generally similar between ETP dolphins and wild bottlenose dolphins from Sarasota, FL.

The only reported deep body temperatures for a *Stenella* species of which we are aware are for captive Hawaiian spinner dolphins (*Stenella longirostris*)(McGinnis *et al.* 1972, Hampton and Whittow 1976). Over a 24-hour period, deep body temperatures of an unknown sex individual, measured with an ingested temperature pill, varied between 37.4-37.9 $^{\circ}$C (McGinnis *et al.* 1972). Mean rectal temperatures of two male and one female spinner dolphins ranged from 37.1-37.3 $^{\circ}$C (Hampton and Whittow 1976). Ten colonic temperatures taken in the ETP study (18.5%) were higher than 37.3 $^{\circ}$C, although temperatures are generally similar to the few measured in captive *Stenella* species.

In the majority of dolphins (72%), deep body temperatures did increase over time when they were held in the processing raft.

Infrared Thermography

We hypothesized that dolphins that had been chased for longer periods of time would have higher surface temperatures relative to the surrounding water temperature than those that had experienced shorter chase times. The infrared thermography results suggest that dolphins that experienced longer total chase times did exhibit higher surface body temperatures (Figure 18) and that those that experienced chases longer than 75 minutes had significantly higher skin surface temperature differentials than those that had undergone shorter chase times (see Figure 17). Because the samples sizes for this analysis were highly variable (see Table 3), and because the data may have included multiple infrared images from the same individual, caution should be exercised when interpreting these results. Both analyses, though, suggest that ETP dolphins that have experienced long chase durations are shunting warm blood to their periphery to dissipate excess body heat to the environment.

Because fishery observers aboard tuna vessels record the chase start time as the time that the speedboats are launched, regression analysis and ANOVAs were used to examine the relationship between speedboat chase time and skin surface temperature. Regression analysis suggested that there was no relationship between skin surface temperature differentials and speedboat chase durations (Figure 20). ANOVAs of binned chase times, though, suggested an apparent significant, although unexpected, effect of

speedboat chase durations on skin surface temperature differentials (Figure 19). Skin temperatures were paradoxically higher after 6-10 and 16-20 minutes, than after 11-15 and 21-25 minutes. Because these analyses did not, by definition, factor in the variable length of the preceding helicopter chase, these results could be an artifact of other factors. There was no apparent relationship between the length of capture time within the net corral and skin surface temperature differentials.

We compared dorsal fin skin surface temperatures of ETP dolphins, taken with the FLIR thermal camera, to those of wild bottlenose dolphins in Sarasota, FL, measured directly with copper-constantan thermocouples. We have infrared thermal images from Sarasota, but the DTIS 500 camera that we utilized during those experiments was not as reliable a quantitative instrument as the FLIR (Pabst *et al.* 2001). In Sarasota, FL, we have measured dorsal fin skin temperatures under two conditions – while dolphins are being held stationary in the net corral and when free-swimming, using the thermal data logger. In Sarasota, the dorsal fin skin temperature measurements in the net corral were taken both when the fin was being held in the air, so the skin surface remained dry, and when the fin was fully submerged. In the ETP, the dorsal fin skin temperatures measured in the net corral were of fins emerging from the water's surface. Thus, a thin film of water enveloped the fin's surface. The temperature of this thin film of water was likely to have been less than that which would have been measured directly at the skin's surface.

In Sarasota, FL, dry dorsal fin temperatures, measured in the net corral, ranged from 28.9-34.7°C; when fully submerged, the dorsal fin skin surface temperatures ranged from 28.7-32.9°C. Mean temperature values recorded from free-swimming dolphins in Sarasota, FL ranged from 30.4-35.0°C. In comparison, the mean dorsal fin temperature of ETP dolphins was approximately 34°C, and ranged from 31.5-38.5°C. Because these fins also were enveloped in a thin film of water, the temperature at the fin was likely to be higher than that imaged with the FLIR. This comparison suggests that dolphins in the ETP, in general, had higher dorsal fin temperatures than bottlenose dolphins in Sarasota, FL. In the ETP study, the skin surface temperatures of dolphins held in the raft were significantly warmer (on average 3.4°C) than those of animals swimming within the net.

Thermal Data Logger Deployments

We hypothesized that dorsal fin heat flux and skin temperature values would be higher in a dolphin during and after a chase (i.e. helicopter over animals) than those recorded before the chase began. We also hypothesized that a dolphin that experienced a prolonged chase would maintain elevated heat flux and skin surfaces temperatures for a prolonged period while in the net corral. The results of the thermal data logger deployments do not fully support these hypotheses.

D227 had higher heat flux values during than before a chase, but significantly decreased heat flux values directly after the chase. D63 displayed high heat flux values both pre- and during the chase, and significantly decreased values directly after the chase. Skin temperature also decreased significantly in D63 post-chase, but this pattern was not observed in D227. These results suggest that both dolphins had or maintained relatively high levels of heat dissipation during the chase, but that this effect was not sustained during the capture time within the net corral. Thus, these two individuals did not appear to require a "cool down" period of sustained high levels of heat dissipation after chase.

Both of these dolphins also did not show elevated deep body temperatures, relative to their first measured temperatures, upon their recapture, although D227's temperature did increase slightly with each successive capture event (see Figure 11).

We had hypothesized that the dolphins might experience high levels of heat flux, skin surface temperatures, and deep body temperatures during prolonged capture time in the net because their ability to swim freely in the net and, thus, to dissipate excess body heat *via* convection, might be decreased. This situation was not the case for either animal, perhaps because there was sufficient room within the net compass for the dolphins to swim freely, particularly for the relatively small groups captured during this study. The thermal effects of being corralled in the net may be different for dolphins that are captured in large groups that are more typical during tuna fishing operations.

While D227 displayed a significant increase in heat flux during chase, D63 did not. We believe that this result may reflect the fact that D63 was already expressing high heat flux levels prior to the time the helicopter began the chase (compare pre-chase heat flux values in Figures 30 and 32). The tracking ship was forced to maintain a very close proximity to the D63 for most of the preceding night since this thermal tag's VHF transmitter had a limited range. Normal following distances were generally kept to at least 1.6 km, but for D63 this distance was likely much less, and the *McArthur* may have been "chasing" D63 long before the capture operations began (see Figure 26, about 0200 on September 20). This situation was improved in the second deployment and the *McArthur*'s following distance was increased for D227.

In the one deployment where speed data were collected, there was no close relationship between heat flux and velocity (see Figure 33). This result was unexpected, because we hypothesized that faster swimming would be associated with both increased rates of heat delivery to the dorsal fin, and increased rates of convective heat loss. No direct relationship existed, though, between velocity and heat flux during chase. This result could be related to a time lag that may exist between increased exercise, increased deep body temperatures and subsequent increases in heat flux.

It is important to note that thermal data logger deployments were only undertaken for two dolphins and, hence, may not be representative of ETP spotted dolphins in general. Because the tag was attached using invasive techniques, it could also be possible that the dorsal fin's thermal responses may have been impacted.

We can, though, compare the heat flux values for D63 and D227 with those collected from wild bottlenose dolphins in Sarasota, FL. In Sarasota, we have measured heat flux at the dorsal fin under two conditions – while dolphins are being held stationary in the net corral, and when free-swimming, using the thermal data logger. While being held stationary, with their fins submerged, dolphins in Sarasota, FL displayed mean heat flux values ranging from 0-225 W/m^2. Mean heat flux values recorded from free-swimming dolphins in Sarasota, FL ranged from 1-322 W/m^2. As a comparison, we also have also measured heat flux across the fin of a captive bottlenose dolphin in Oahu, HI. This dolphin displayed pre-exercise heat flux values ranging from 10-25 W/m^2 and post-exercise heat flux values between 190-290 W/m^2. Thus, heat flux values measured from ETP dolphins fall into the range of values that we have measured in wild and captive bottlenose dolphins (Figure 34).

SUMMARY

The results of this portion of the CHESS study suggest the following interpretation of the thermal response of dolphins to chase and capture. A dolphin's thermal response begins during the helicopter phase of the chase. Chased dolphins appear to respond by increasing the rate of heat dissipation to the environment. Prolonged chase times (75 minutes or longer) result in measurably higher skin surface temperatures of dolphins. These elevated surface temperatures suggest relatively high rates of heat delivery to the skin's surface *via* blood flow. Dolphins that have experienced chases less than 75 minutes in length do not appear to maintain elevated levels of heat flux and high skin surface temperatures once the chase ends and the animals are within the net corral. This result may be, in part, because the animals in this study did not appear to experience any impediment to free-swimming, and, thus, to convective heat dissipation, within the net corral. Extended chase durations and capture times within the net were not statistically associated with elevated deep body temperatures in this study. One of the 48 individuals, though, displayed an unusually high body temperature of $37.9^{o}C$. Because there was no evidence from blood diagnostic tests to confirm illness, physiological or exertional stress is the more likely explanation of this individual's elevated body temperature.

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Table 2. Dolphins handled for deep core temperature measurements during ETP CHESS operations. Gender determination remains unresolved for animals with a ? in the sex category. Note that core temperatures are recorded 2 minutes after probe insertion (* temperature recorded 8 minutes after probe insertion due to mechanical error).

SET #	Animal ID #	Sex	Total Length (cm)	Time 1st Handled	Time of Ist Core Temp	Ist Core Temp (°C)
1	D12	F	181	13:11	13:17	36.4
1	D14	?	191	13:48	13:53	37.4
2	D17	?	180	10:31	10:34	35.2
2	D18	M	164	10:39	10:44	36.5
2	D23	M	213	11:06	11:08	37.6
3	D24	F	194	10:29	10:33	37.0
3	D25	F	196	10:38	10:41	36.9
6	D26	F	174	10:53	10:56	36.5
6	D29	F	185	10:54	10:57	36.7
6	D30	F	183	11:08	11:11	37.0
6	D33	?	170	11:10	11:14	36.8
6	D34	F	204	11:20	11:21	36.8
7	D37	F	174	10:47	10:50	36.7
7	501/D34	F	204	10:49	10:52	36.7
7	D32	F	186	10:55	10:58	36.6
7	D38	M	167	10:58	11:02	37.1
8	D39	F	189	16:06	16:09	37.2
8	D31	F	171	16:08	16:11	37.5
8	502/D29	F	185	16:15	16:23	36.9
8	D36	M	177	16:17	16:21	37.0
8	D40	M	161	16:26	16:29	37.4
8	D45	M	188	16:26	16:28	37.2
9	D41	M	209	9:14	9:19	36.7
9	D46	M	205	9:16	9:19	36.9
11	D47	F	185	15:13	15:21	34.8
11	506/D29	F	185	15:14	15:17	36.8
20	D42	M	188	14:56	15:00	37.0
21	505/D42	M	188	15:05	15:11	36.5
21	D48	F	172	15:06	15:09	36.7
21	D49	M	170	15:12	15:15	36.5
21	D60	F	205	15:14	15:17	37.5
21	D59	F	206	15:18	15:22	36.2
22	504/D203	F	191	9:55	9:59	36.5
22	508/D193	?	181	9:56	9:59	37.3
22	D65	F	192	10:06	10:09	37.0
22	D61	F	188	10:13	10:17	37.9
23	D62	?	190	10:06	10:10	37.0
23	D66	F	189	10:07	10:10	36.8
23	D67	M	202	10:13	10:18	36.8
23	D63	M	190	10:17	10:21	37.5
23	D68	M	178	10:19	10:23	37.5
24	512/D67	M	202	9:50	9:53	37.8*
24	511/D63	M	190	9:51	9:56	37.3
25	D227	M	204	13:26	13:31	37.2
25	D221	?	195	13:26	13:29	37.3
25	D222	M	200	13:36	13:39	36.6
25	D228	M	202	13:43	13:48	37.1
25	D224	F	193	13:48	13:51	36.8
27	513/D227	M	204	16:31	16:37	37.1
27	515/D244	M	208	16:38	16:42	36.9
27	517/D245	M	197	16:39	16:42	36.9
27	516/D257	M	202	16:45	16:52	37.0
27	D225	M	200	16:47	16:51	37.2
27	D229	M	200	16:58	17:01	37.5
27	D230	F	196	17:03	17:07	37.5

Bivariate Fit of without saran wrap By with saran wrap
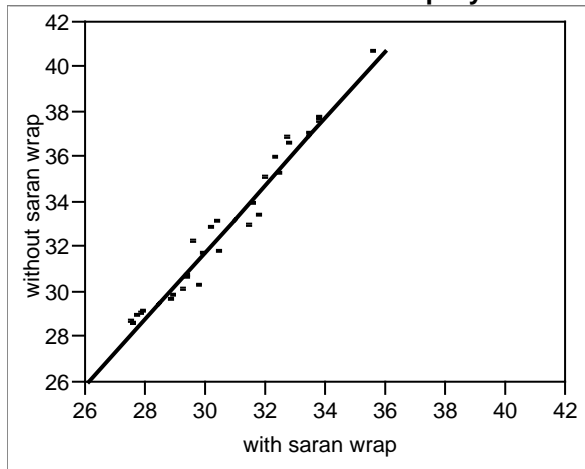
———Linear Fit

Linear Fit

without saran wrap = -13.13789 + 1.4969785 with saran wrap

Summary of Fit

RSquare	0.966714
RSquare Adj	0.965481
Root Mean Square Error	0.627049
Mean of Response	32.66759
Observations (or Sum Wgts)	29

Analysis of Variance

Source	DF	Sum of Squares	Mean Square	F Ratio
Model	1	308.31619	308.316	784.1397
Error	27	10.61614	0.393	Prob > F
C. Total	28	318.93233		<.0001

Parameter Estimates

Term	Estimate	Std Error	t Ratio	Prob>|t|
Intercept	-13.13789	1.639903	-8.01	<.0001
with saran wrap	1.4969785	0.053459	28.00	<.0001
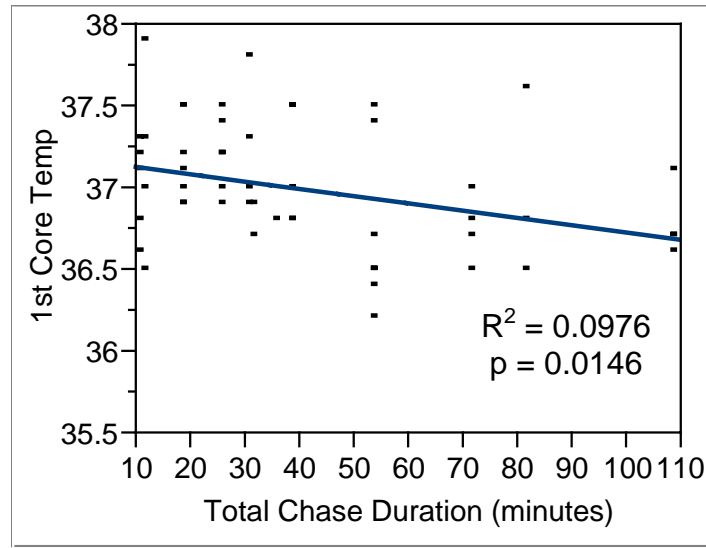
Figure 4. First core temperatures (^{o}C) recorded from all dolphins against the total chase duration. Four dolphins were excluded from this analysis: D17 and D47, the two outliers, 512/D67 wh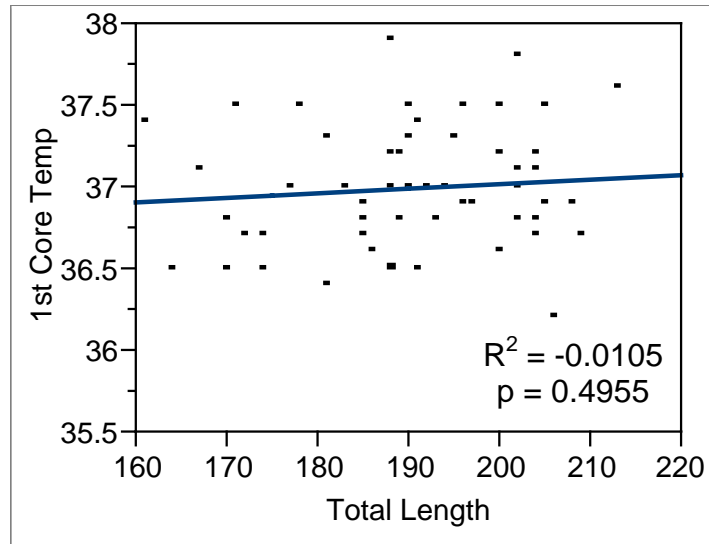ose temperature was taken at 8 minutes after probe insertion, and D42 for whom there was no chase start time (i.e. the time the helicopter was over dolphins).
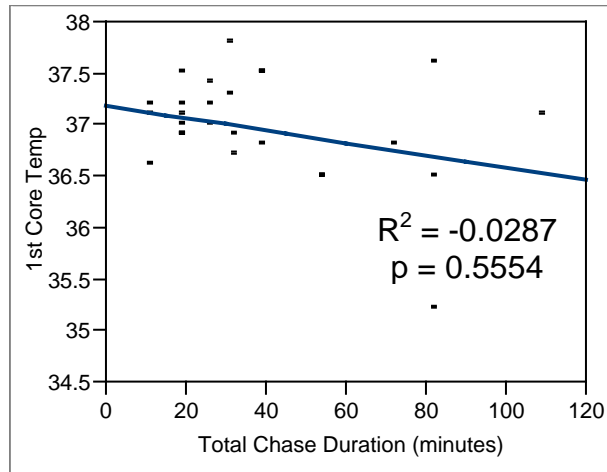
Figure 5a. First core temperatures (^{o}C) recorded from male dolphins against the total chase duration. Three dolphins were excluded from this analysis: D17, the outlier, 512/D67 whose temperature was taken at 8 minutes after probe insertion, and D42 for whom there was no chase start time (i.e. the time the helicopter was over dolphins).
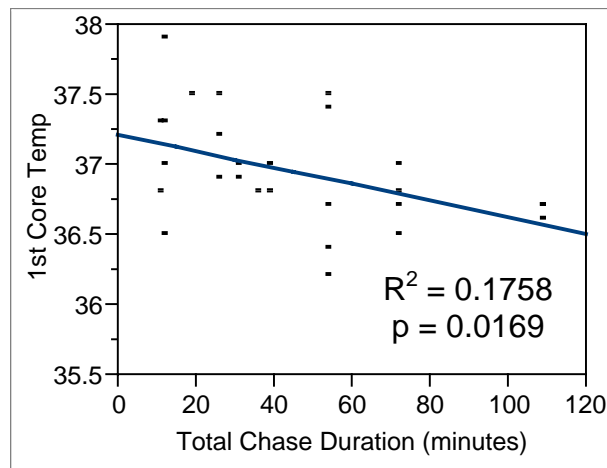
Figure 5b. First core temperatures (^{o}C) recorded from female dolphins against the total chase duration. The outlier D47 was excluded from this analysis.
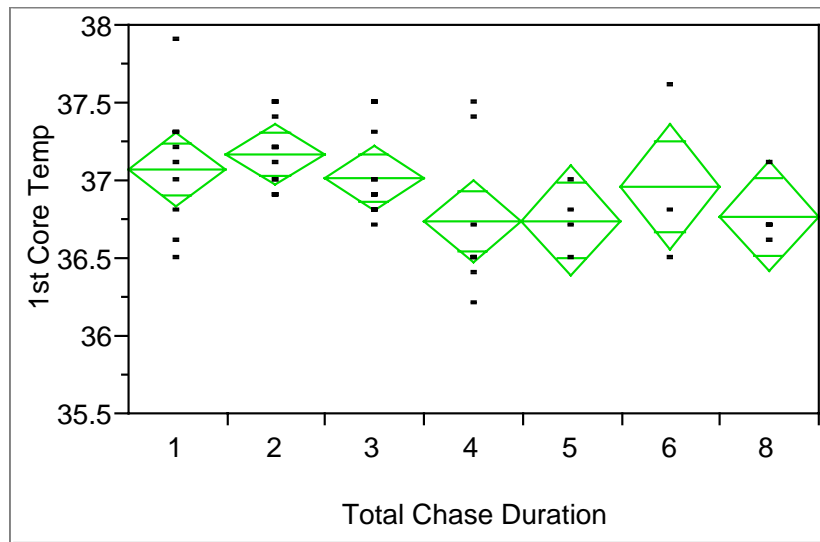
Figure 6. First core temperatures (^{o}C) recorded from all dolphins against the total chase duration (see Table 1 for chase duration codes). ANOVA revealed no detectable relationship between deep body temperature and total chase duration. The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Four dolphins were excluded from this analysis: D17 and D47, the two outliers, 512/D67 whose temperature was taken at 8 minutes after probe insertion, and D42 for whom there was no chase start time (i.e. the time the helicopter was over dolphins).

Figure 7a. First core temperatures (^{o}C) recorded from male dolphins against the total chase duration (see table 1 for chase duration codes). ANOVA revealed no detectable relationship between deep body temperature and total chase duration. The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Four dolphins were excluded from this analysis: D17 and D47, the two outliers, 512/D67 whose temperature was taken at 8 minutes after probe insertion, and D42 for whom there was no chase start time (i.e. the time the helicopter was over dolphins).

Figure 7b. First core temperatures (^{o}C) recorded from female dolphins against the total chase duration (see table 1 for chase duration codes). ANOVA revealed no detectable relationship between deep body temperature and total chase duration. The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Four dolphins were excluded from this analysis: D17 and D47, the two outliers, 512/D67 whose temperature was taken at 8 minutes after probe insertion, and D42 for whom there was no chase start time (i.e. the time the helicopter was over dolphins).

$$R^2 = 0.0115$$
$$p = 0.5307$$

Figure 8. Core temperature (^{o}C) at 2 minutes after probe insertion for all dolphins against the amount of time spent in the net (in minutes). Three dolphins were excluded from this analysis: D17 and D47, the two outliers, and 512/D67 whose temperature was taken at 8 minutes after probe insertion.
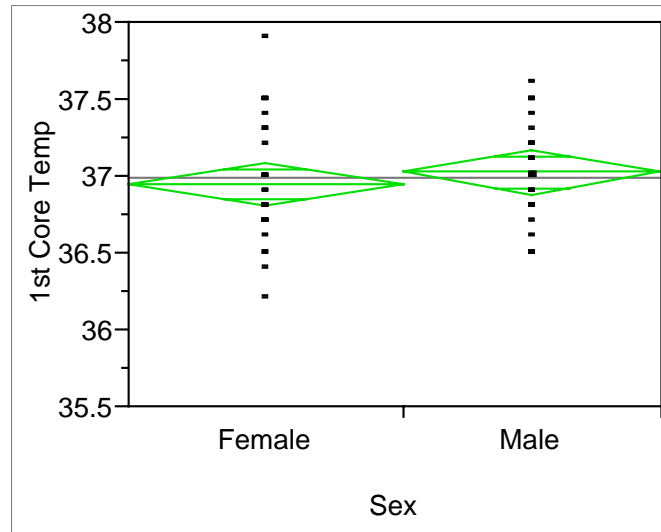
Figure 9. Female *vs.* male deep core temperatures (°C). Core temperatures were all taken at 2 minutes after probe insertion. There was no difference between mean deep body temperatures of males and females. The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Three dolphins were excluded from this analysis: D17 and D47, the two outliers, and 512/D67 whose temperature was taken at 8 minutes after probe insertion.

Figure 10. Core temperatures taken 2 minutes after probe insertion (^{o}C) recorded from all dolphins against total body length (cm). Three dolphins were excluded from this analysis: D17 and D47, the two outliers, and 512/D67 whose temperature was taken at 8 minutes after probe insertion. Separate analyses for each sex also showed no significant relationships between core temperature and body length.
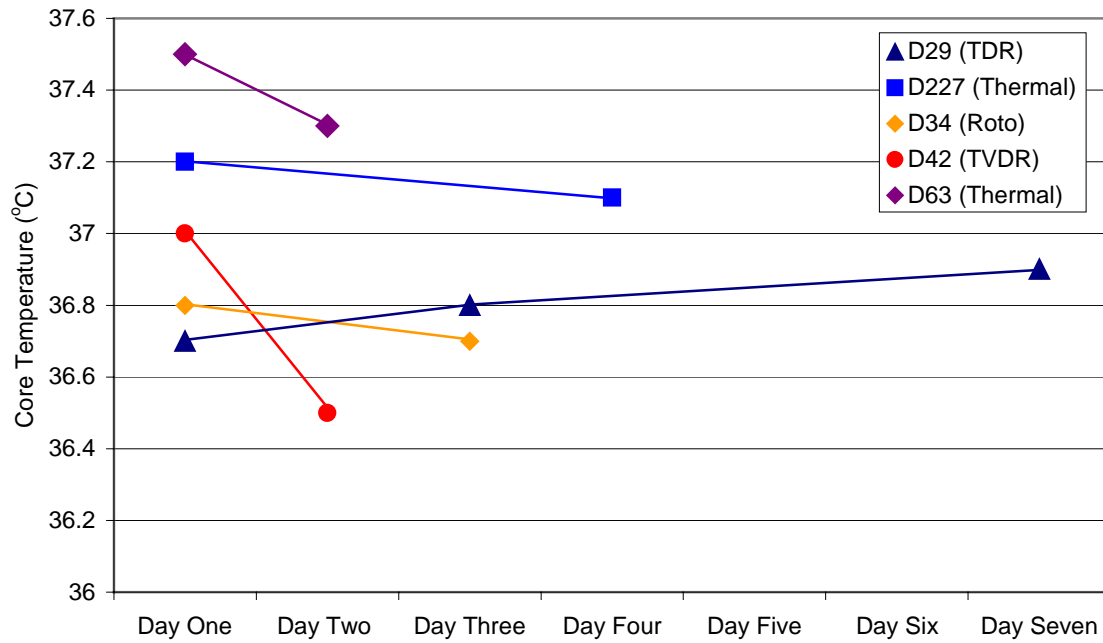
Figure 11. Serial deep body temperatures (taken two minutes after probe insertion) of recaptured dolphins. Day one represents the first capture event, while following days are the number of days after the first capture that the dolphins were recaught. D67 was excluded from this analysis because its recapture temperature was taken at 8 minutes after probe insertion.
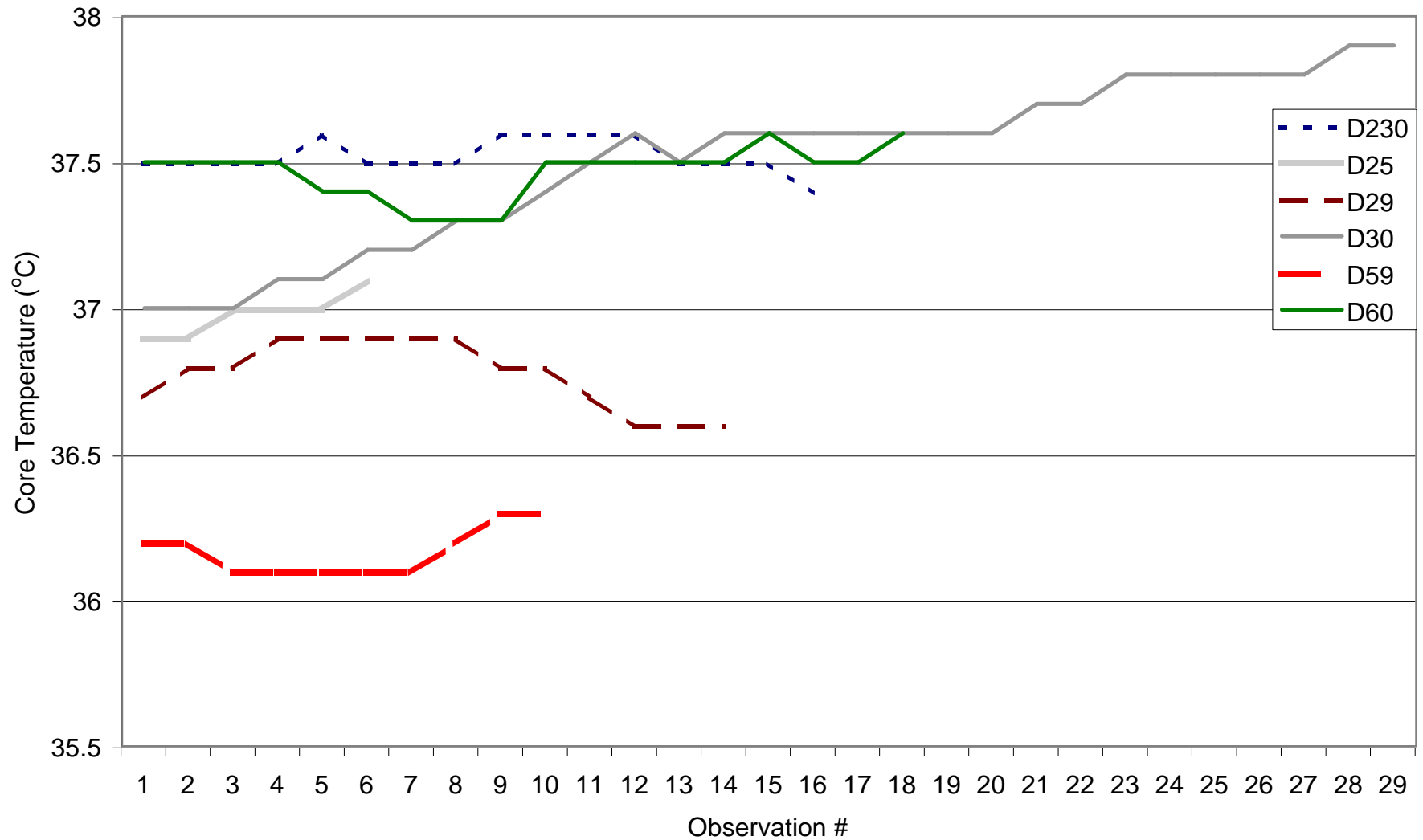
Figure 12. Examples of female deep body temperatures recorded over time in the raft. Each line represents a single individual (see legend). Note that observation #1 is two minutes after probe insertion and the following observations are at 30 second intervals.
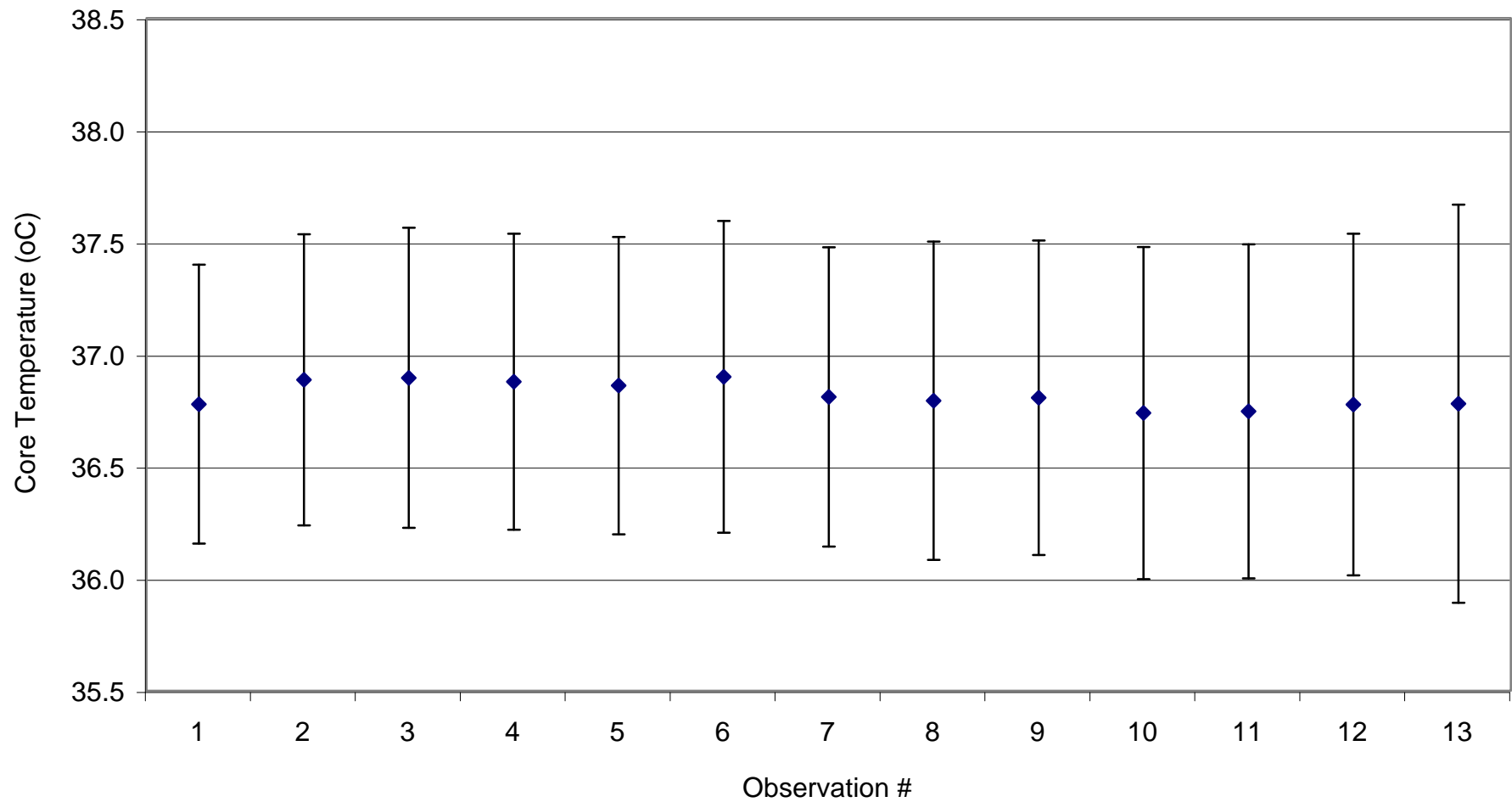
Figure 13. Mean core temperatures and standard deviations for females recorded over time in the raft. Note that observation #1 is two minutes after probe insertion and the following observations are at 30 second intervals. The sample size of female deep core temperatures decreases from 23 individual temperatures at observation #1 to 10 individual temperatures at observation #13.
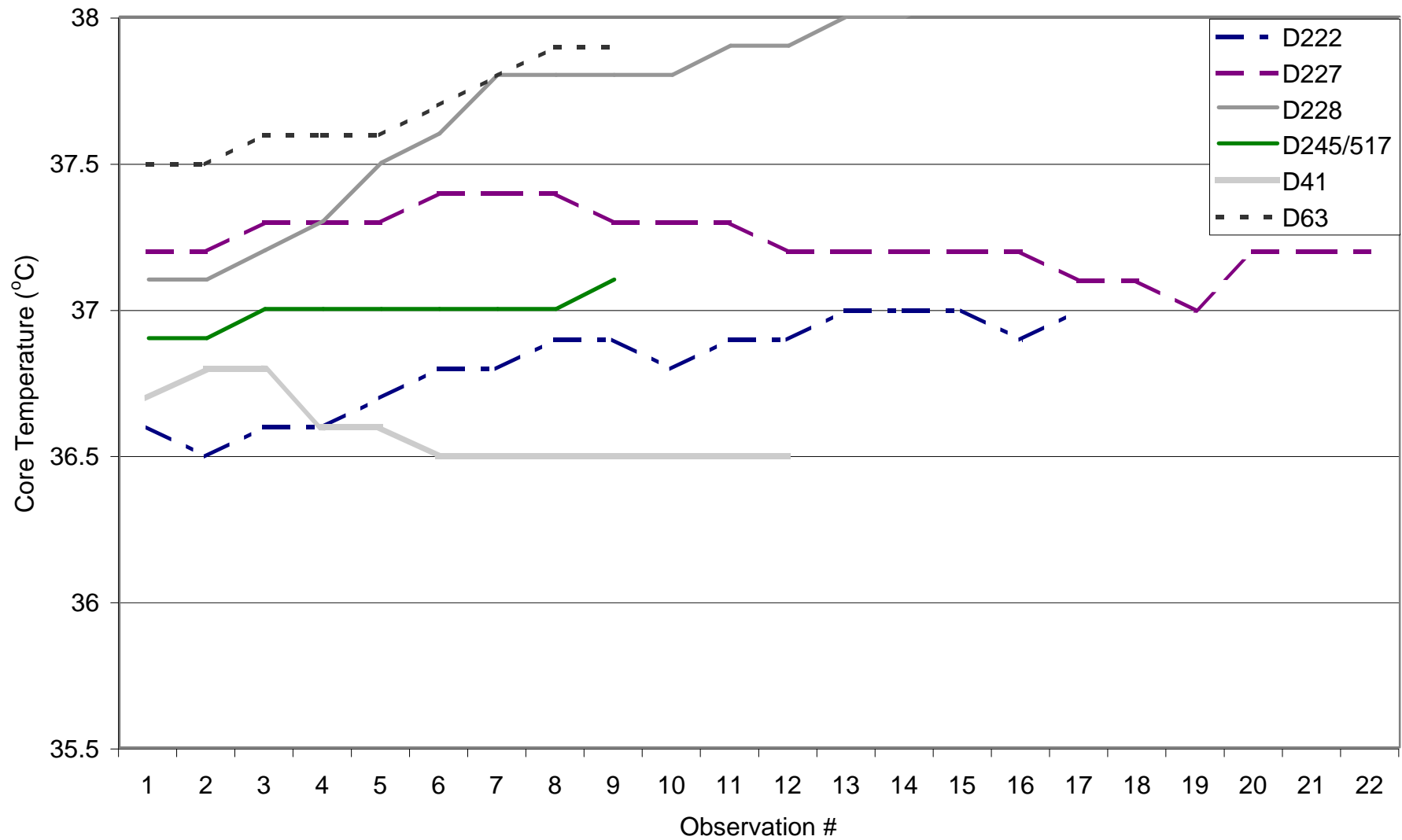
Figure 14. Examples of male deep body temperatures over time in the raft. Each line represents a single individual (see legend). Note that observation #1 is two minutes after probe insertion and the following observations are at 30 second intervals.
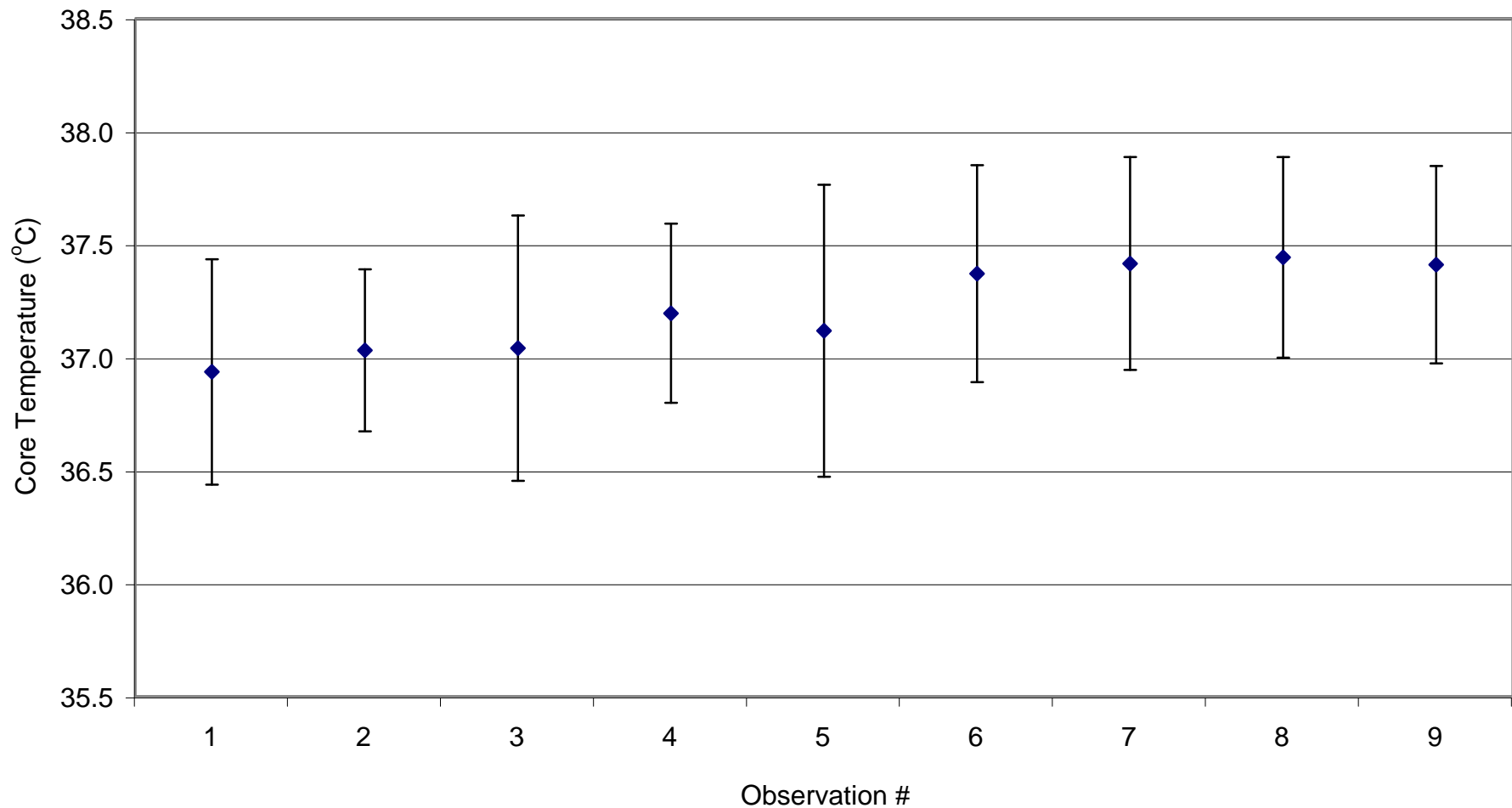
Figure 15. Mean core temperatures and standard deviations for males recorded over time in the raft. Note that observation #1 is two minutes after probe insertion and the following observations are at 30 second intervals. The sample size of male deep core temperatures decreases from 23 individual temperatures at observation #1 to 11 individual temperatures at observation #9.
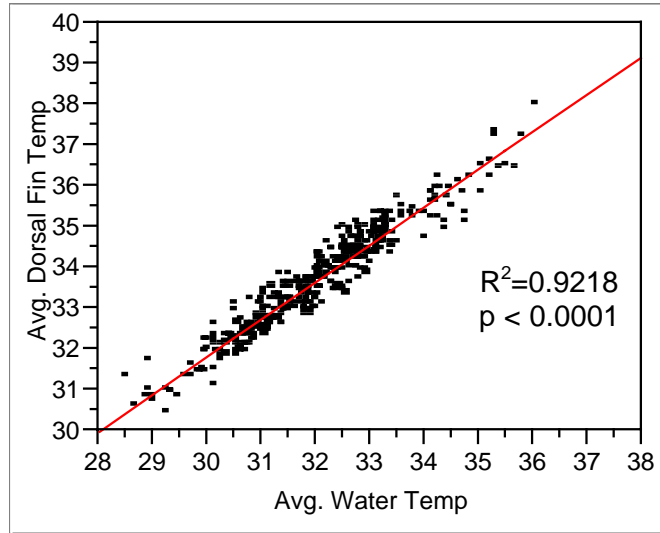
Figure 16a. Average dorsal fin surface temperature against average water surface temperature (^{o}C) (n=422). The equation for the linear fit is:
average dorsal fin temperature = 4.2160801 + 0.9188283 average water temperature
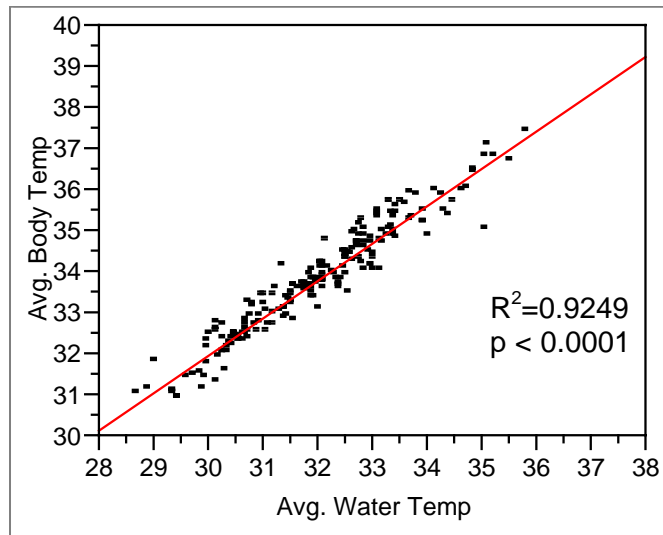
Figure 16b. Average body surface temperature against average water surface temperature (^{o}C) (n=202). The equation for the linear fit is:
average body temperature = 4.7033945 + 0.9088819 average water temperature
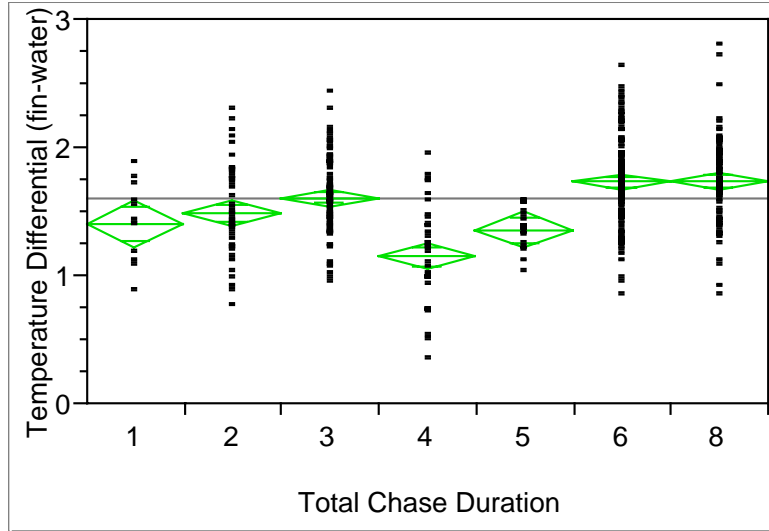
Figure 17a. Temperature differential (T_{fin}-T_{water}) in ^{o}C against total chase duration (see Table 1 for chase duration codes). The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Chase durations 6 and 8 have significantly higher mean temperature differentials than all other chase durations except 3. Chase duration 3 has a significantly higher mean temperature differential than chase durations 4 and 5. Chase duration 7 is absent due to n=0.
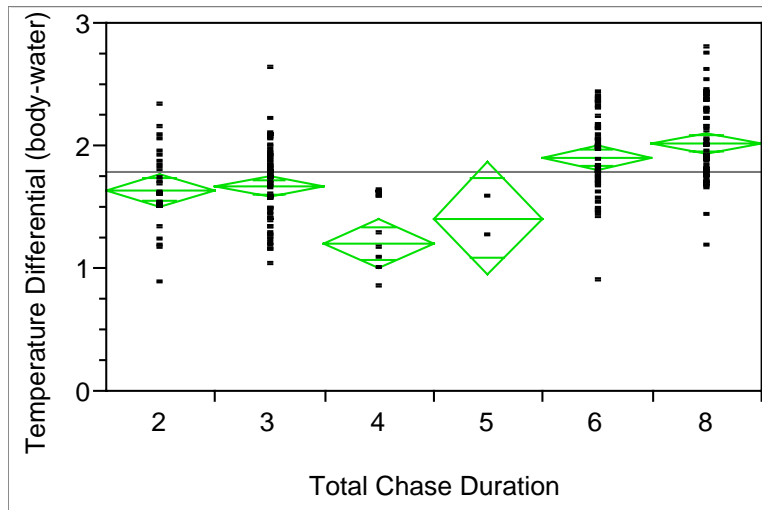
Figure 17b. Temperature differential (T_{body}-T_{water}) in ^{o}C against total chase duration (see Table 1 for chase duration codes). The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Chase durations 6 and 8 have significantly higher mean temperature differentials than chase durations 2, 3 and 4. Chase duration 5 is not significantly different from any other chase duration, probably due to small sample size (n=2). Chase durations 1 and 7 are absent due to n's=0.
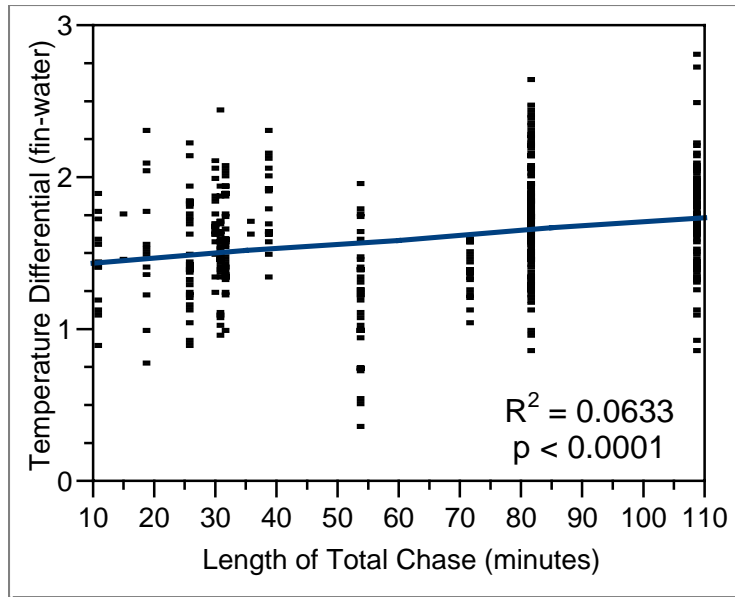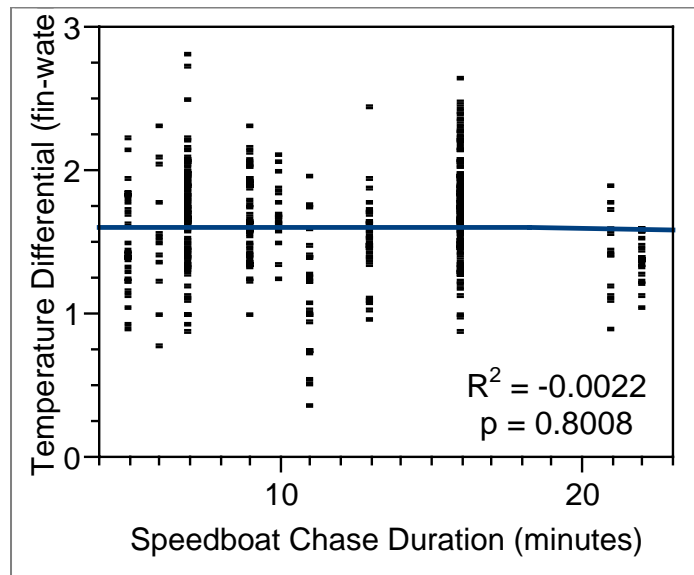
Figure 18a. Temperature differential (T_{fin}-T_{water}) in ^{o}C against the total chase duration in minutes.
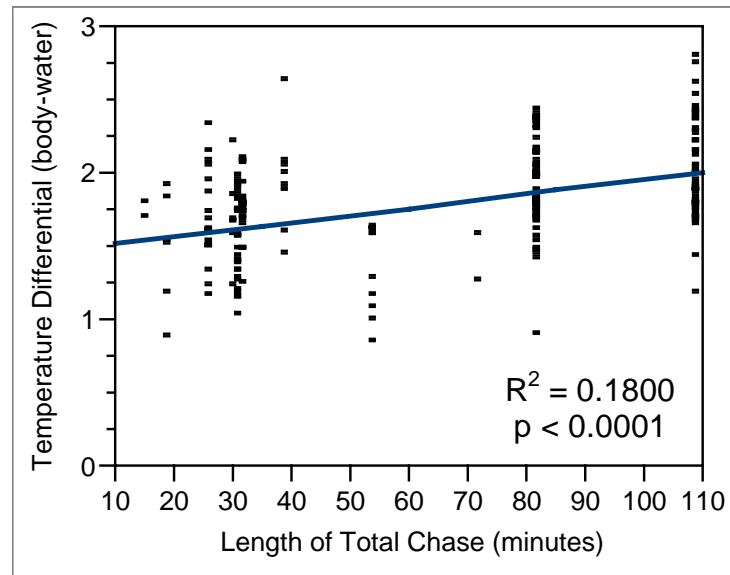
Figure 18b. Temperature differential (T_{body}-T_{water}) in ^{o}C against the total chase duration in minutes.
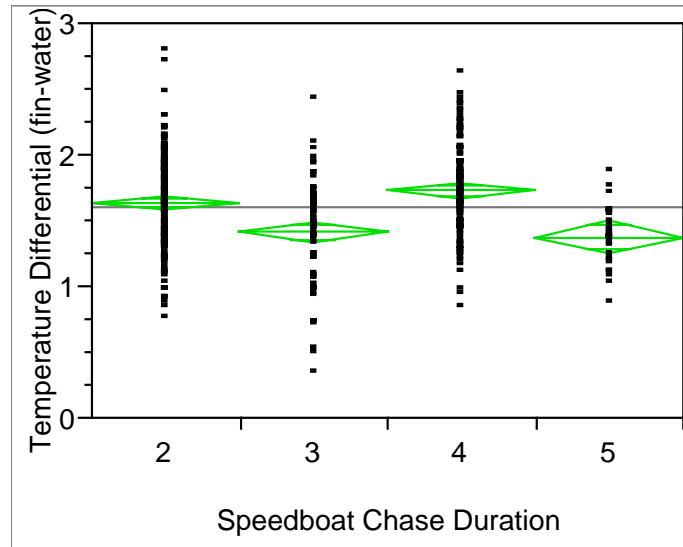
Figure 19a. Temperature differential (T_{fin}-T_{water}) in ^{o}C against speedboat chase times (see Table 1 for chase duration codes). The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Chase durations 2 and 4 have significantly higher mean temperature differentials than chase durations 3 and 5. Chase duration 1 is absent due to n=0.
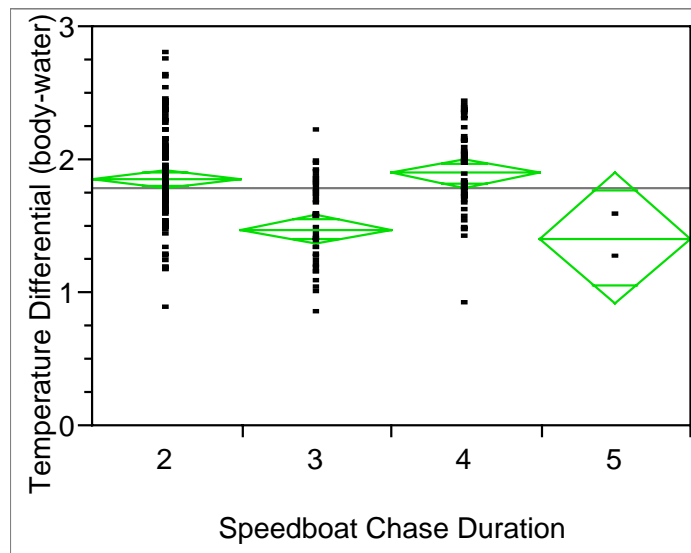
Figure 19b. Temperature differential (T_{body}-T_{water}) in ^{o}C against speedboat chase times (see Table 1 for chase duration codes). The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Chase durations 2 and 4 have significantly higher mean temperature differentials than chase duration 3. Chase duration 5 is not significantly different from any other chase duration, probably due to small sample size (n=2). Chase duration 1 is absent due to n=0.

Figure 20a. Temperature differential ($T_{fin} - T_{water}$) in ^{o}C against the speedboat chase duration in minutes.
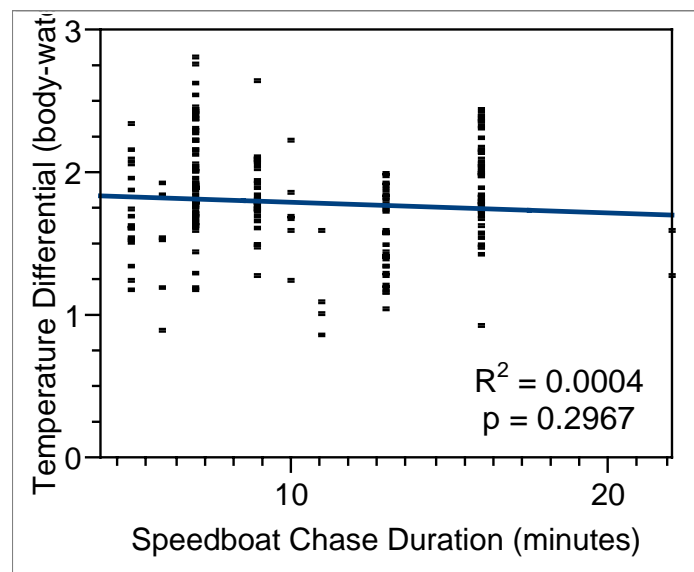
Figure 20b. Temperature differential ($T_{body} - T_{water}$) in ^{o}C against the speedboat chase duration in minutes.
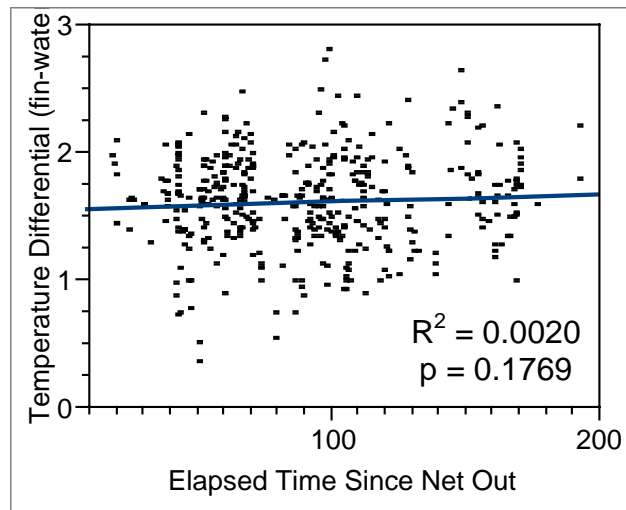
Figure 21. Temperature differentials (^{o}C) for the dorsal fin against elapsed time since net out (minutes).
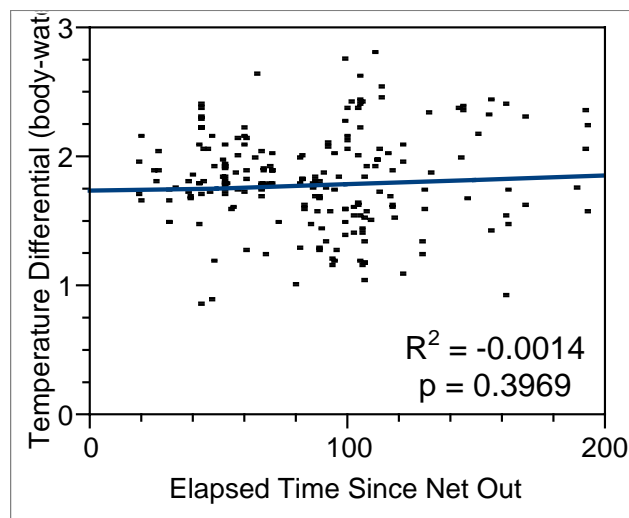
Figure 22. Temperature differentials (^{o}C) for the body against elapsed time since net out (minutes).
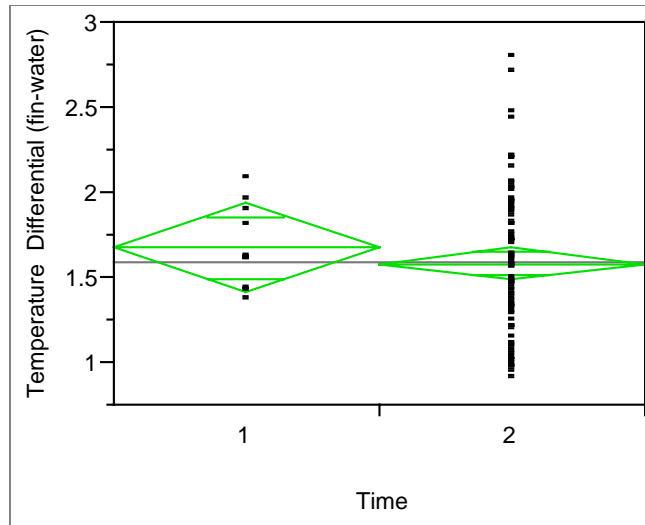
Figure 23a. Temperature differential (^{o}C) against the extreme time blocks of the capture process. The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Time code 1 represents thermal images taken within 30 minutes of net out. Time code 2 represents thermal images taken within 20 minutes of backdown complete. There are no significant differences of mean temperature differentials across these two time blocks.
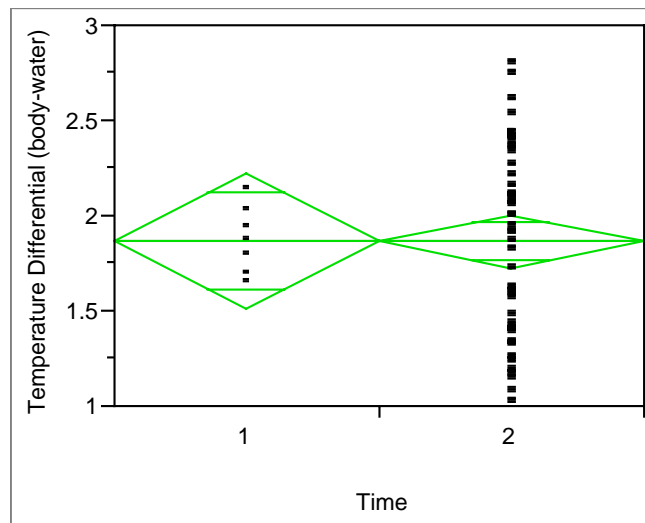
Figure 23b. Temperature differential (^{o}C) against the extreme time blocks of the capture process. The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Time code 1 represents thermal images taken within 30 minutes of net out. Time code 2 represents thermal images taken within 20 minutes of backdown complete. There are no significant differences of mean temperature differentials across these two time blocks.
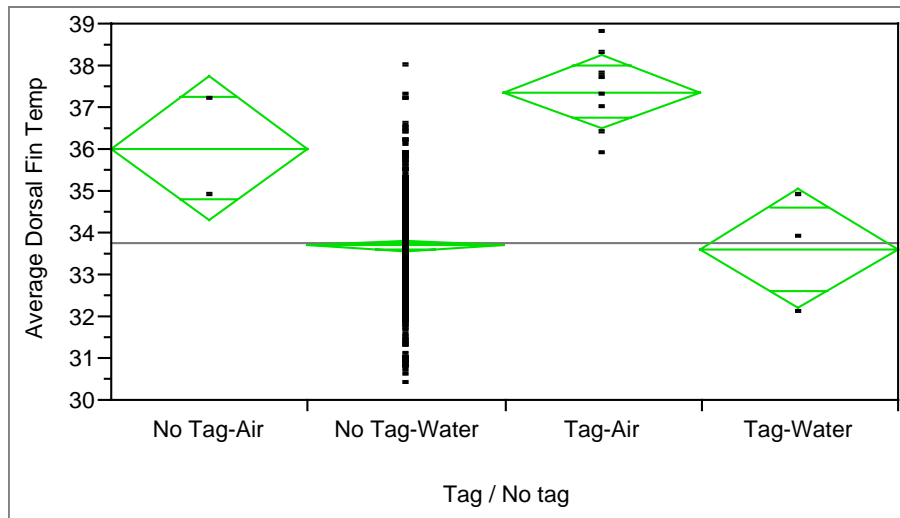
Figure 24a. Average dorsal fin temperatures (^{o}C) for tagged and untagged fins in water and air. Fins in air represent images taken while the dolphins were held in rafts. The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Tagged and untagged fins in water are not significantly different from each other. Likewise, tagged and untagged fins in air are similar. Thus, in both water and in air all tagged and untagged fins were pooled for further analyses (see Figure 24b).
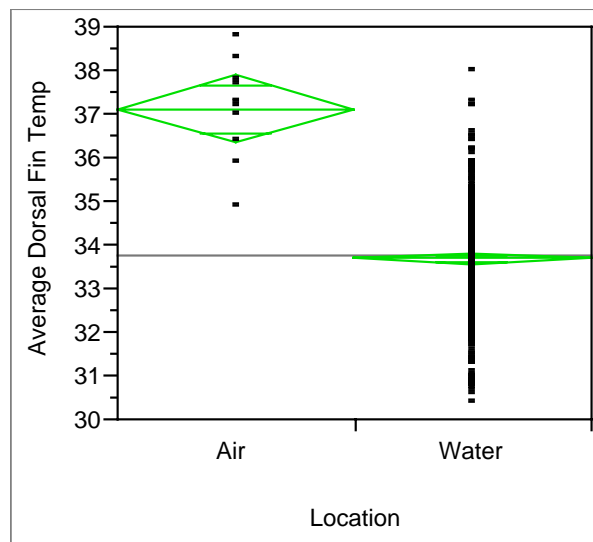
Figure 24b. Average dorsal fin temperatures (^{o}C) for fins in air and water. The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Fin temperatures measured in air are significantly different than fin temperatures measured in water and are on average 3.4 ^{o}C warmer.
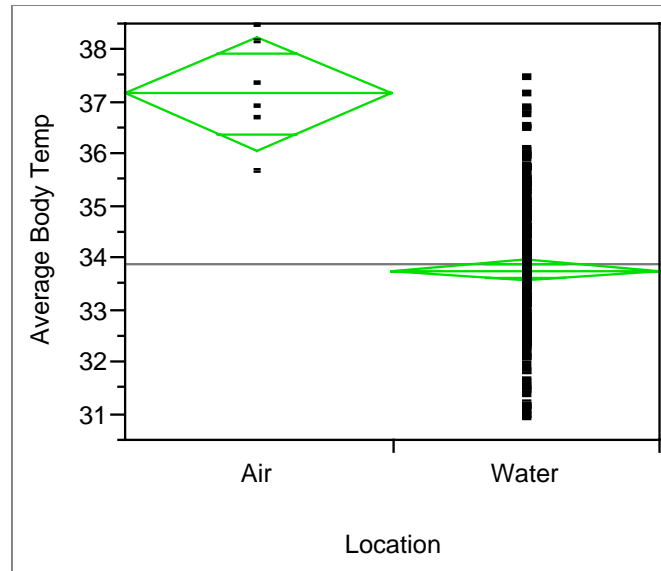
Figure 25. Average body temperatures (^{o}C) for animals in air and water. The gray diamonds show the mean and 95% confidence interval for each group. The center lines represent the group means, while the top and bottom of the diamonds form the 95% confidence intervals for the means. Body temperatures measured in air are significantly different than body temperatures measured in water and are on average 3.4 ^{o}C warmer.
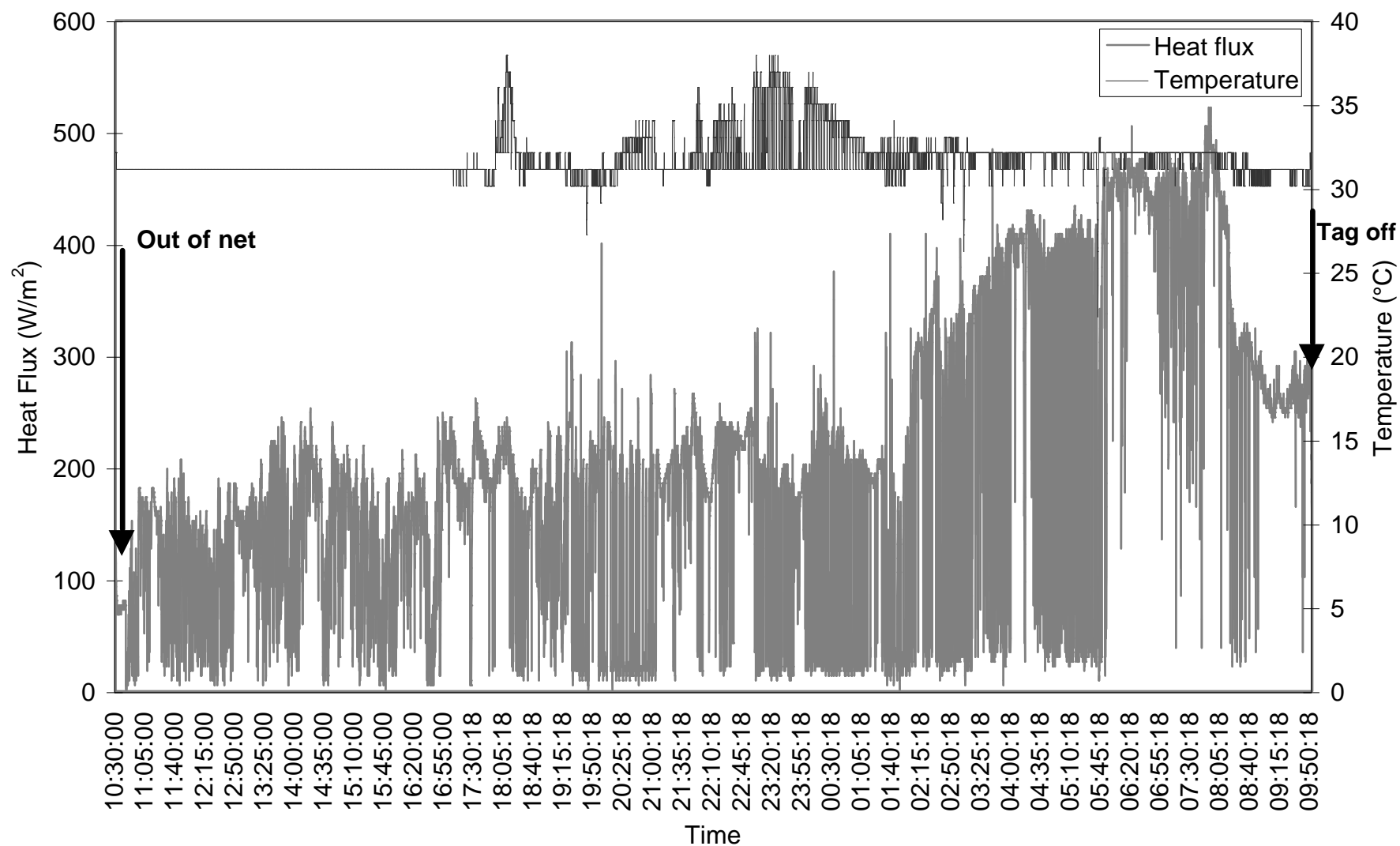
Figure 26. Entire thermal data logger record for D63, September 19-20.
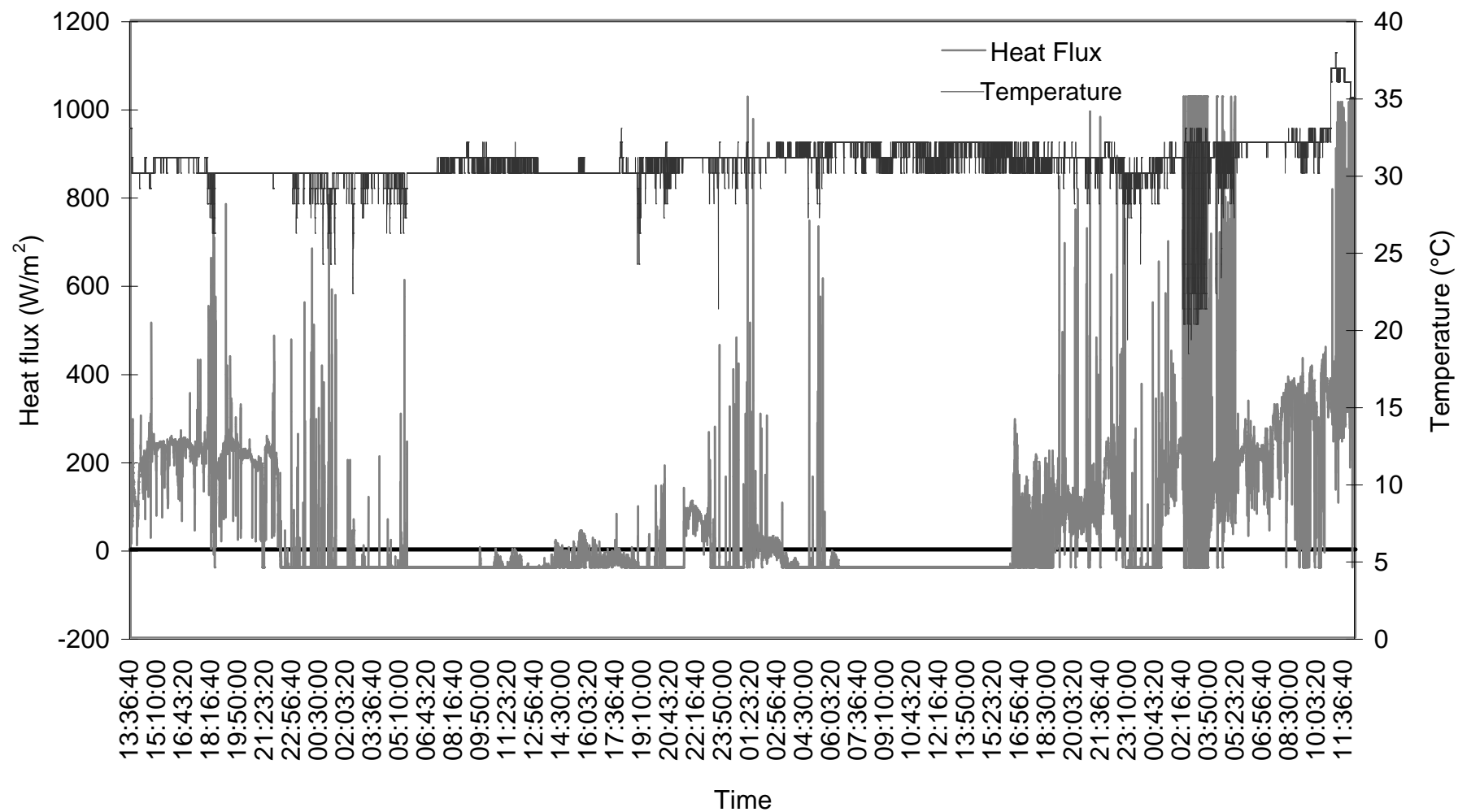
Figure 27. Entire thermal data logger record for D227, September 22-25. Note that the heat flux record contains long time segments during which the disc was not collecting data.
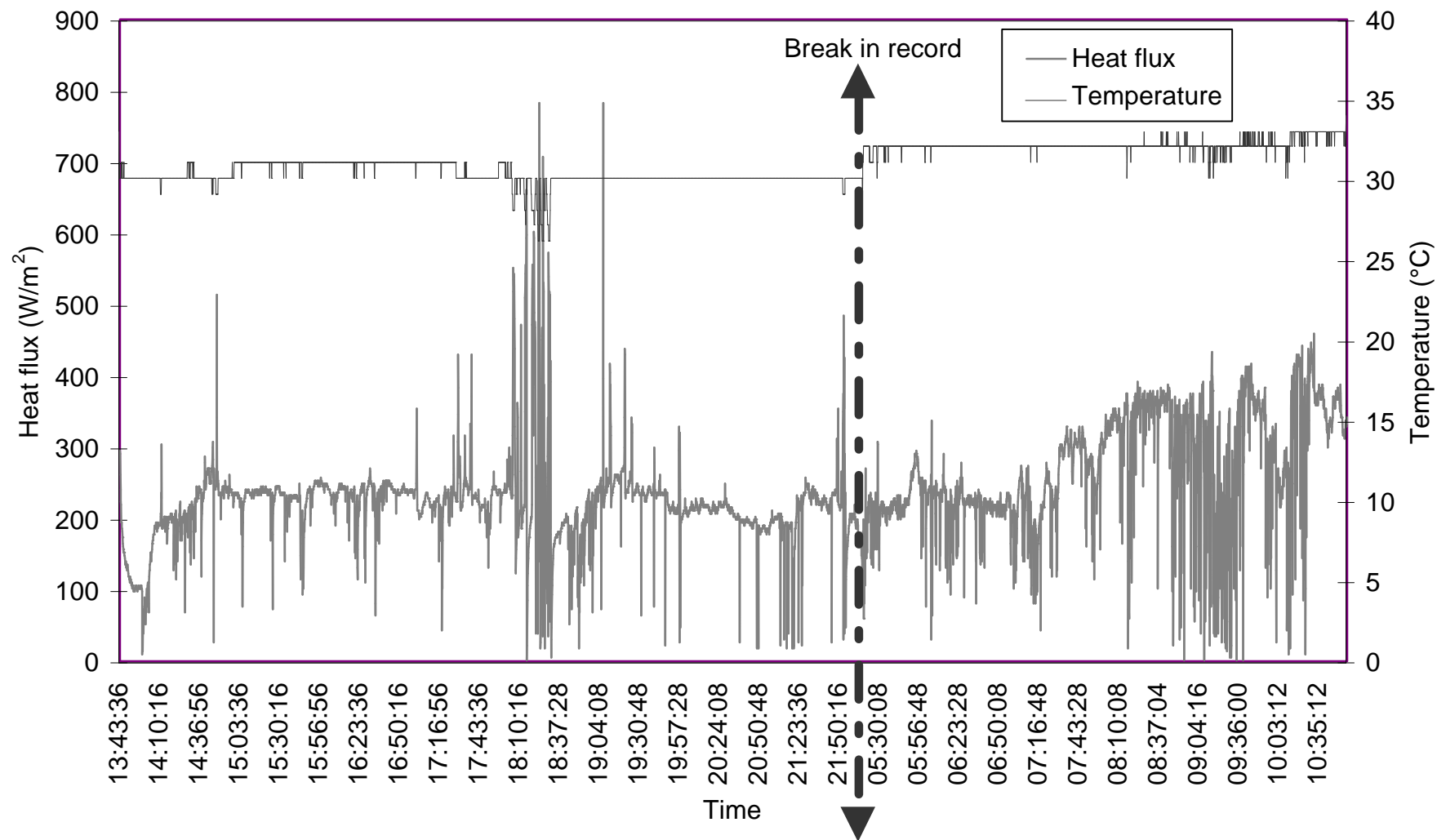
Figure 28. Edited thermal data record for D227 excluding period of malfunction. Initial data segment is for September 22; final data segment is for the first, unsuccessful capture attempt on September 25.
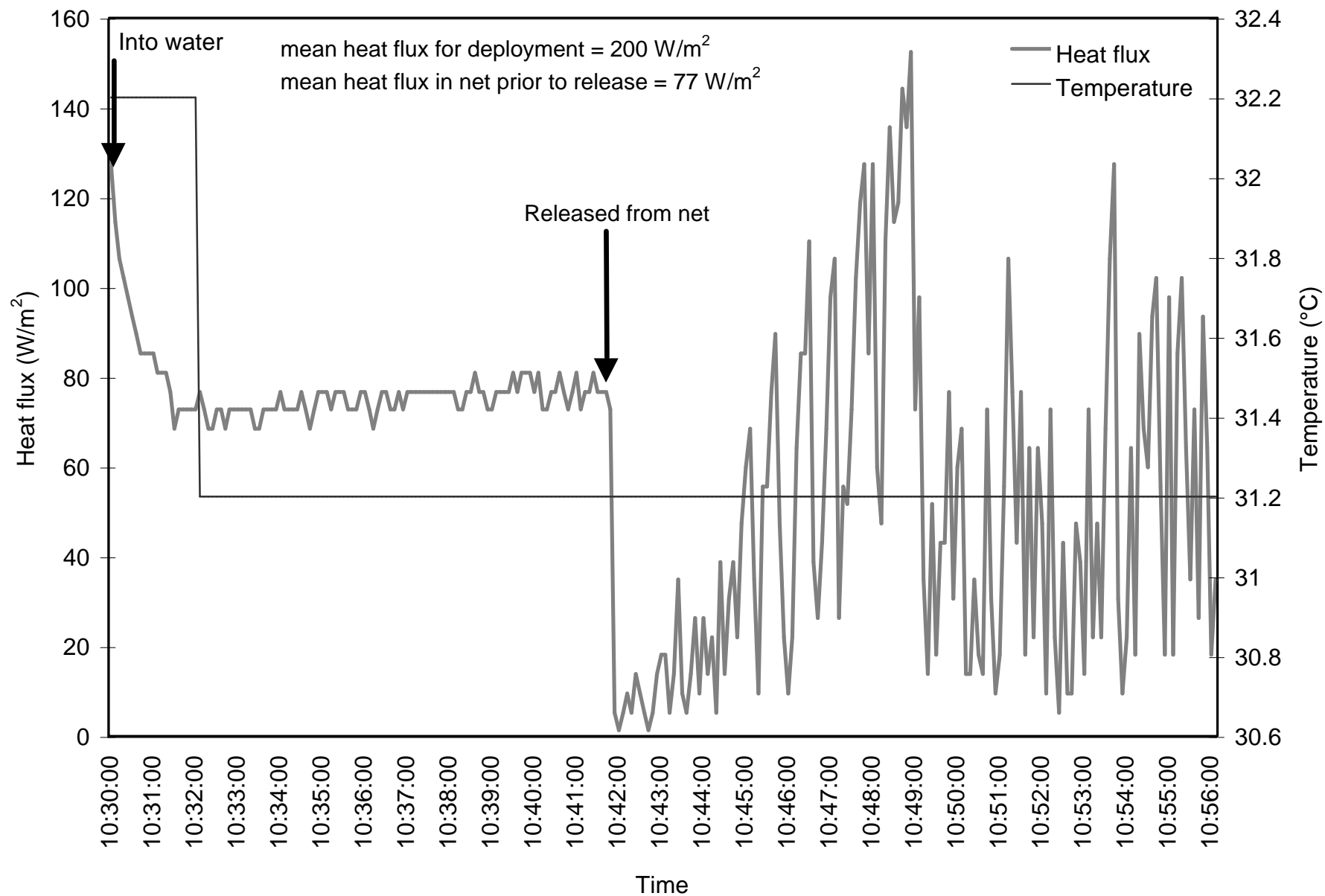
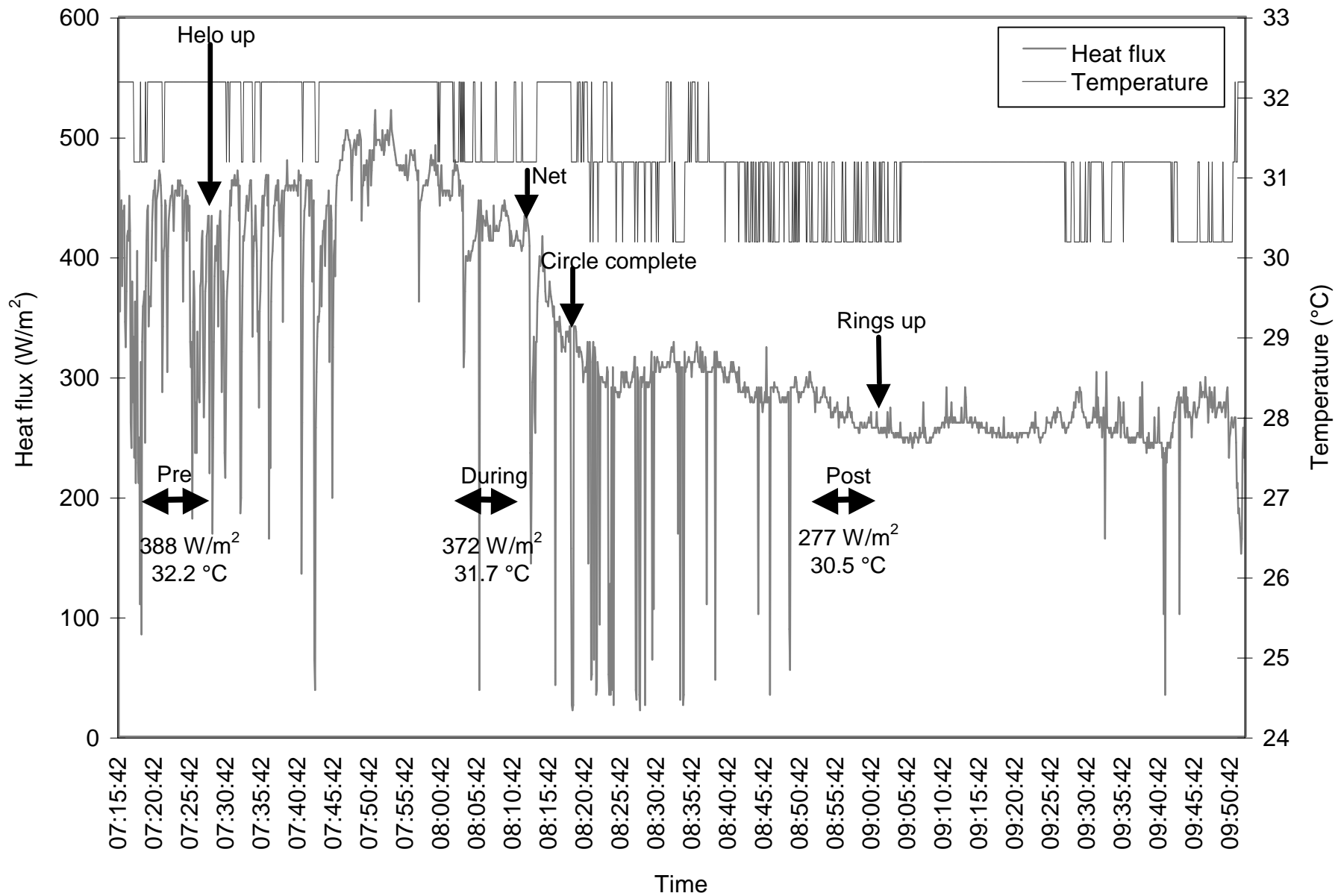**Figure 29.** Thermal data logger record for D63 for first half hour after release from the processing raft.

Figure 30. Thermal data logger record for D63 during chase and recapture on September 20.
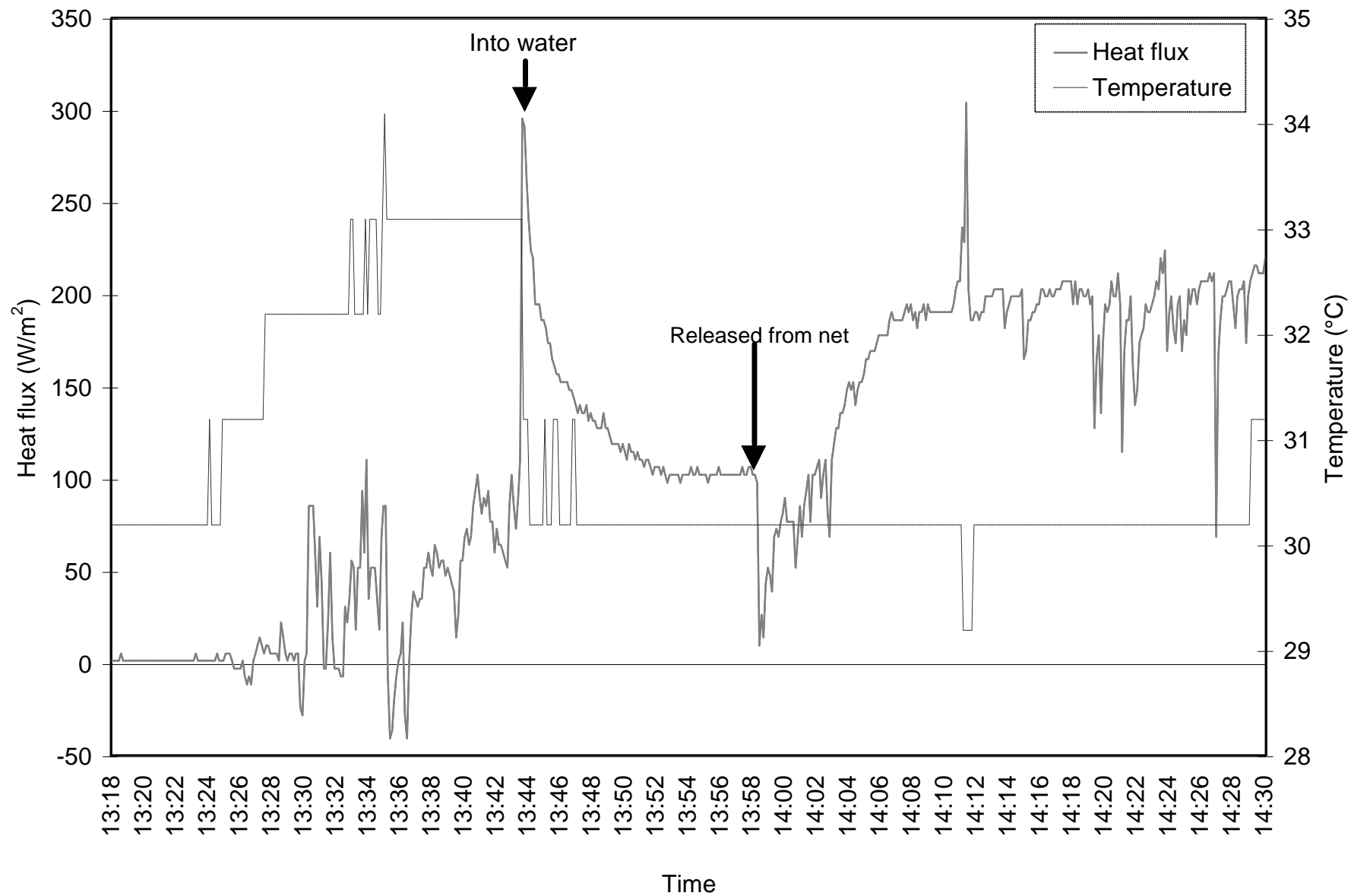
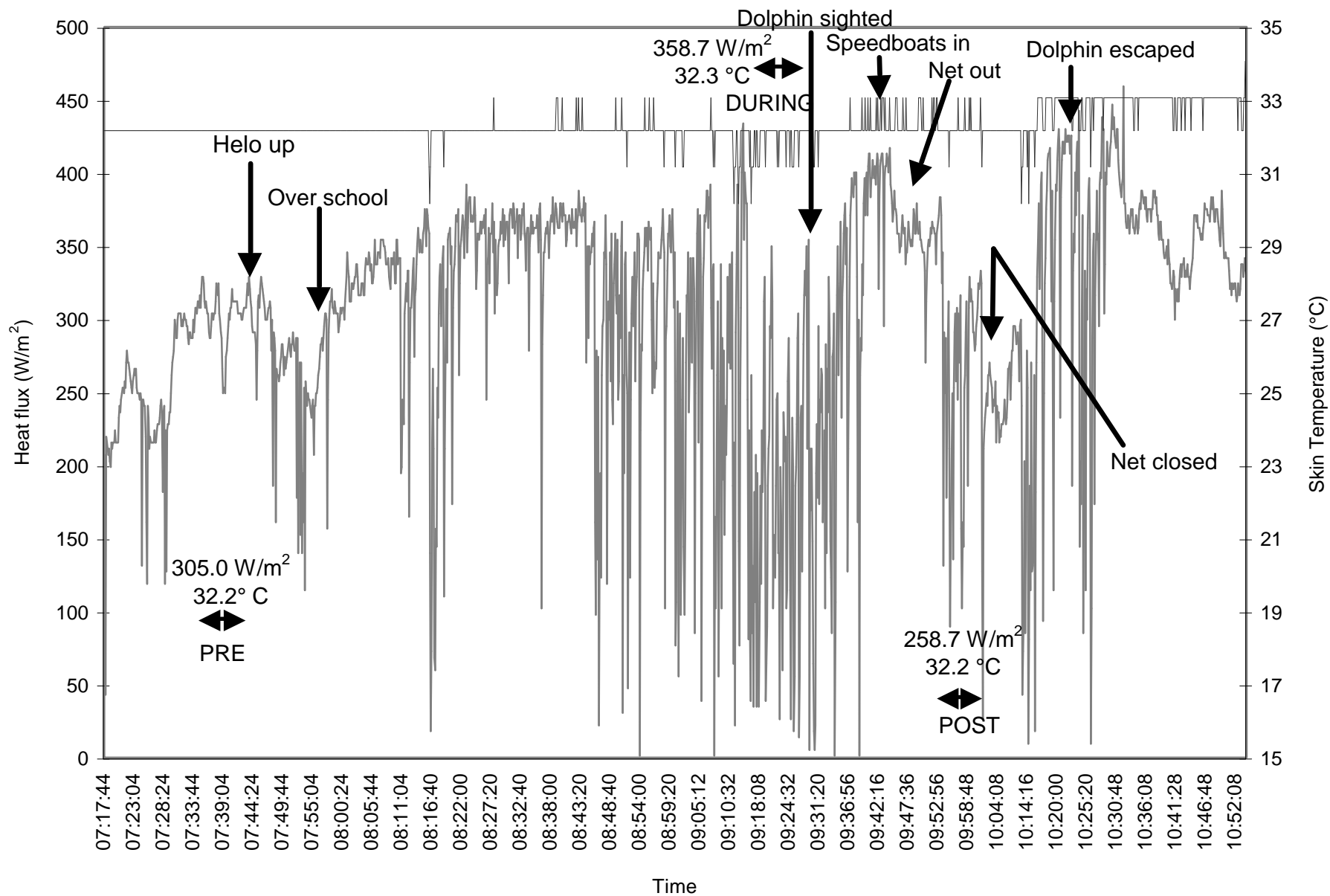**Figure 31.** Thermal data logger record for D227 for first 45 minutes after release from the processing raft.

Figure 32. Thermal data logger record for D227 during the first chase and recapture attempt.
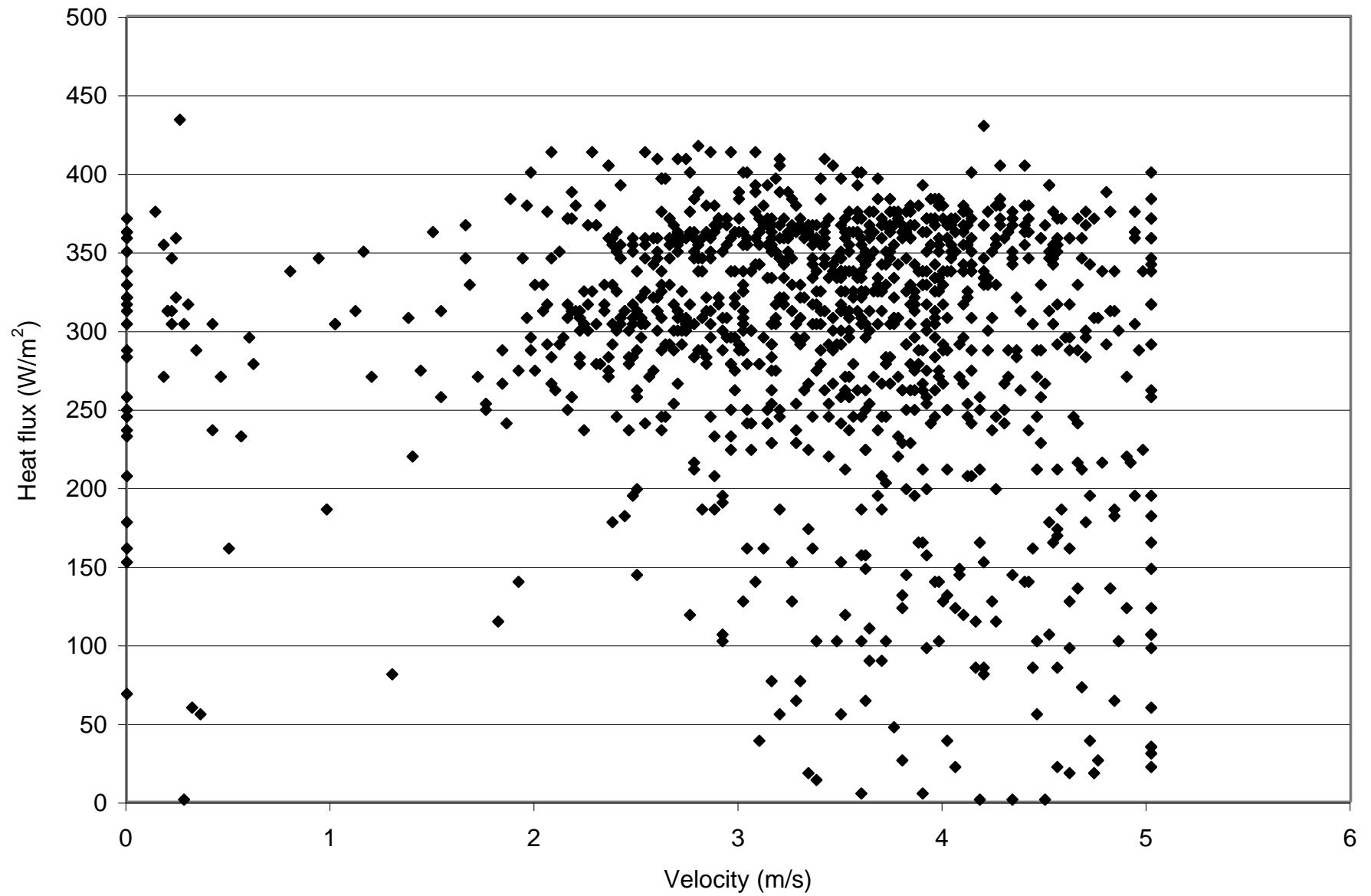
Figure 33. Relationship between heat flux and swimming speed, as measured by the Mark 8 tag on D227, during the unsuccessful chase and recapture event recorded from 07:40 - 10:20.
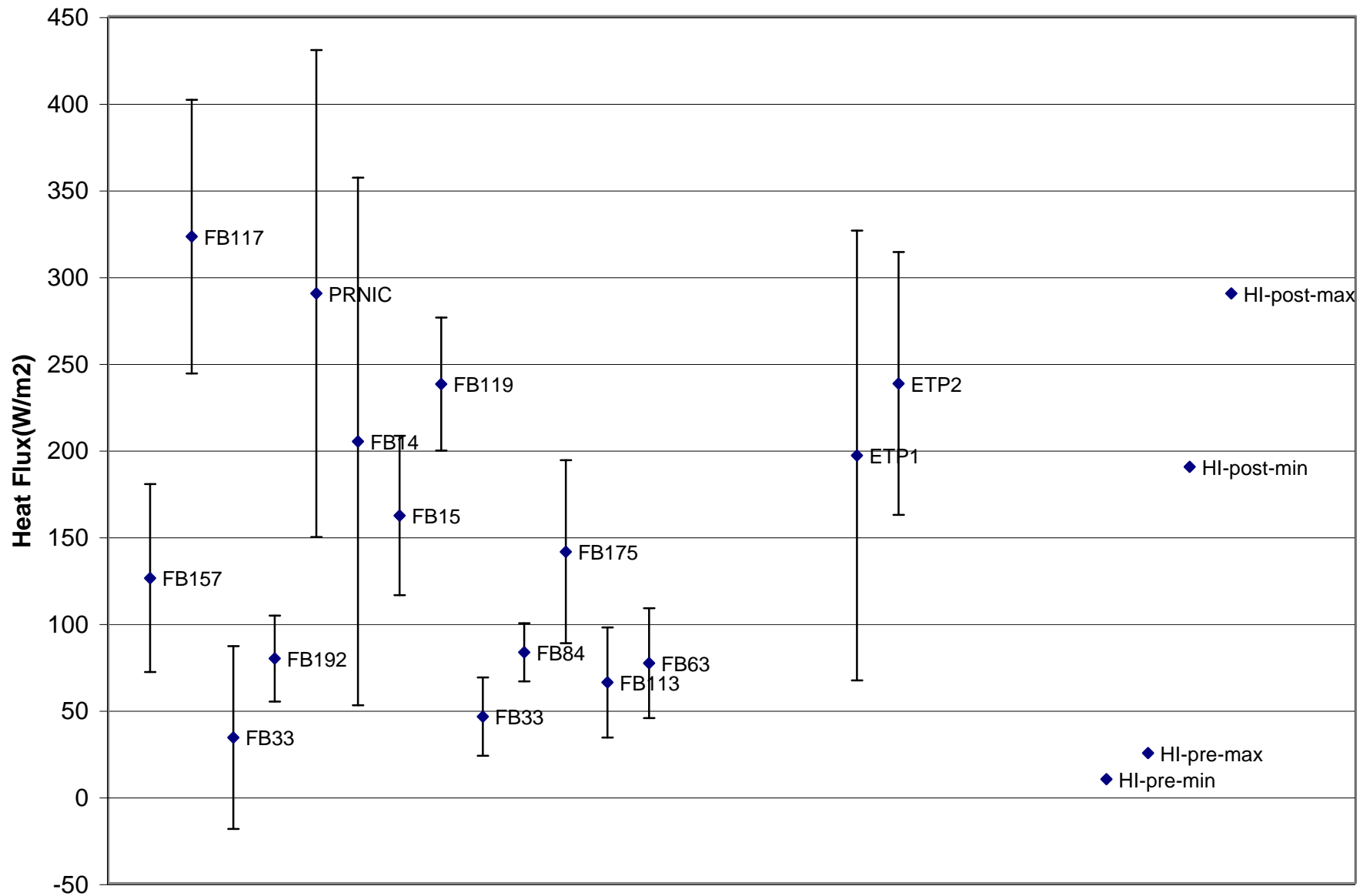
Figure 34. Mean and range of heat flux values for Sarasota dolphins (FB #) and ETP dolphins and minimum and maximum heat flux values for a captive dolphin (HI) pre- and post-exercise.

APPENDIX

Responses to reviewer comments   26 April 2002

Attached please find the revised text for: "Measuring Temperatures and Heat Flux from Dolphins in the Eastern Tropical Pacific: Is Thermal Stress Associated with Chase and Capture in the ETP-Tuna Purse Seine Fishery?".

The revisions include the following:

(1) addition of a Table of Contents
(2) addition of Executive Summary
(3) addition of statement that core body temperatures may be biased by handling, as suggested by Dr. Greg Bossart (page 17)
(4) addition of a statement that D61, the animal with the highest deep core temperature, had also undergone extended chase times within the net, as suggested by Dr. Michael Scott (page17).

We are not re-sending any of the tables, figures and graphs, because these have not changed.

We were very pleased with the positive reviews that the manuscript received, and we would like to extend our sincere appreciation to Dr.'s Bossart, De Guise, Ortiz, Martineau, and Mann for their careful review of our project. The comments of Dr. Martineau were very insightful, and we will continue to analyze the in-net data to investigate the relationship between depth, water temperature and heat flux in corralled animals. We discovered in Sarasota that dolphins do achieve higher heat flux rates by diving to depths where water temperatures are lower (results presented at the 14th Biennial of the Society for Marine Mammalogy meetings, 2001). Thus, even in shallow bays, depth specific temperature profiles can influence heat flux. This will be an exciting area of future data analysis.